Performance evaluation in shipboard test of the Auramarine ballast water management systems CrystalBallast®250

Auramarine Ltd.

Report

June 2012

This report has been prepared under the DHI Business Management System certified by DNV and specifically for ballast water management system testing certified by Lloyd's Register	
Quality Management	BWMS Testing
ISO 9001	IMO Resolution MEPC.174(58) Annex part 2
Quality Management System certified according to DS/EN ISO 9001 by Det Norske Veritas, Business Assurance, Danmark A/S	Lloyd's Register

Approved by
04-06-2012 X Approved by Signed by: Jens Tørsløv

Performance evaluation in shipboard test of the Auramarine ballast water management systems CrystalBallast®250

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M/S Ahtela in Hundested Harbour

Project No	11811494
Classification	Confidential

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Abbreviations

Abbreviation	Description
AVG	Average
BWMS	Ballast water management system
CFU	Colony-forming units
DOC	Dissolved organic carbon
DNV	Det Norske Veritas
DPM	Disintegrations per minute
DWT	Deadweight tonnage
FR	Field replicate
IMO	International Maritime Organization
MEPC	Marine Environment Protection Committee
MPN	Most probable number
NTU	Nephelometric turbidity units
POC	Particulate organic carbon
PSU	Practical salinity units
QAPP	Quality assurance project plan
QMP	Quality management plan
SOP	Standard operating procedure
STD	Standard deviation
TSS	Total suspended solids
UVT	UV transmittance

1 Executive summary

DHI provides independent verification testing services to developers of ballast water management systems. DHI's quality assurance project plan is consistent with the requirements of the International Convention for the Control and Management of Ships Ballast Water and Sediments.

From November 2011 through May 2012, DHI conducted shipboard tests of the Auramarine ballast water management system CrystalBallast®250 (CB250) in accordance with DHI's certification by Lloyd's Register verified by Det Norske Veritas (DNV). The ability of the CB250 to (a) successfully treat ballast water without interruption and (b) meet the IMO D-2 ballast water discharge standard was evaluated during a series of six valid test cycles.

All six test cycles were conducted in Hundested Harbour, where the source water can be characterized as brackish water (15-25 practical salinity units) with a high diversity of planktonic organisms.

The average densities of viable organisms in the ≥50 µm size class varied from approx. 3,500 to approx. 29,000 organisms per m^3 in the inlet water. For the size class ≥10 and <50 µm, the average densities in inlet water varied from approx. 110 to 400 organisms/mL when determined by inverted microscopy. The contents of *E. coli* in the inlet water were generally low and enterococci were hardly detected. The inlet water concentrations of organisms ≥50 µm and the smaller planktonic organisms (≥10 and <50 µm) fulfilled the validity criteria defined in the IMO G8 guidelines. For shipboard testing, there are no requirements in the IMO G8 guidelines in relation to the density of bacteria in the inlet water.

The numbers of viable organisms in the ≥50 µm size class in the treated discharge water were 0 per m^3 in three test cycles and 0.20-1.9 per m^3 in the remaining three test cycles, which is 5-50 times below the threshold value defined in the IMO D-2 standard. The density of viable algae in the treated discharge water was determined to be <0.18-0.48 organisms/mL by use of a most probable number (MPN) assay, and micro-zooplankton ≥10 and <50 µm was not observed in the treated discharge water. By using the results of the MPN assay, the density of viable organisms representing the ≥10 and <50 µm size class in the treated discharge water was thus 20-55 times below the IMO D-2 standard. Measurements of the primary production showed a decrease of 99-100% after treatment in the ballast water management system compared with the control with untreated ballast water, which confirmed that the treatment resulted in an immediate impact on the algal photosynthesis. In the treated discharge water the average contents of *E. coli* and enterococci were below the detection limit in all test cycles. *Vibrio cholerae* was not identified in any of the test cycles.

The CB250 system functioned properly during all six test cycles and was highly effective at reducing live organism densities fulfilling the IMO consistent challenge conditions. Live organisms in the size classes defined in the IMO G8 guidelines were discharged at densities below the IMO D-2 standard.

2 Introduction

The objective of this project was to conduct a shipboard test of the Auramarine CrystalBallast®250 in accordance with the guidance given in Resolution MEPC.174(58), Guidelines for approval of ballast water management systems (G8) (IMO 2008), hereafter referred to as the IMO G8 guidelines.

DHI holds a certificate of compliance issued by Lloyd's Register. The acting classification society for the shipboard test of the Auramarine ballast water management system (BWMS) CrystalBallast®250 (CB250) was DNV (Det Norske Veritas).

DHI has no involvement, intellectual or financial, in the mechanics, design or marketing of the BWMS whose performance has presently been evaluated. To ensure that DHI tests are uncompromised by any real or perceived individual or team bias relative to test outcomes, DHI test activities are subject to rigorous quality assurance, quality control procedures and documentation.

During the shipboard testing campaigns, a combined 75 m^3/h + 250 m^3/h CrystalBallast® ballast water treatment unit with two different filtration technologies and two different UV reactor systems was tested. The CB250 consisted of one UV reactor and only one of the two filters was used during the ballast operations in each test cycle.

During six consecutive valid test cycles, the CB250 was evaluated for its ability to: (a) successfully treat ballast water without interruption and (b) meet the IMO D-2 standard (IMO 2004) at discharge.

3 Testing laboratory

DHI is an independent, international consulting and research organisation with the objectives to advance technological development and competence within the fields of water, environment and health. DHI established the DHI Ballast Water Centre with the purpose to provide performance evaluation of BWMS. The DHI Ballast Water Centre includes land-based test facilities and environmental laboratories in Denmark and Singapore.

The shipboard test was carried out by:

DHI
Agern Allé 5
DK-2970 Hørsholm
Denmark

4 Ballast water management system

A description of the CB250 as provided by Auramarine is included in the quality assurance project plan (QAPP) in Appendix C.

Three of the six test cycles (#1, #4 and #5 in Table 5.1) were conducted with one filter type, FilterSafe BSF – 100H (40 µm) whereas the three remaining test cycles (#2, #3 and #6 in Table 5.1) were conducted with another filter type, Boll & Kirch 6.18.2 (30 µm). Specifications of the filters and the UV intensity readings from the CB250 system are available in the data logging provided in Appendix A.

5 Experimental design

5.1 Trial periods and locations

The shipboard test was conducted on-board the trailer carrier M/S Ahtela (IMO 8911736). M/S Ahtela is a cargo ship from 1991 and has been converted in 1998 and 2008. M/S Ahtela is a DNV class general cargo carrier Ro-Ro registered in Rauma, Finland. The vessel has a deadweight tonnage (DWT) of 6,600 tons and a gross tonnage of 8,610 tons. During the shipboard testing period the M/S Ahtela was not in regular route. Therefore, ballast and de-ballast operations were conducted 3-4 times per week every other week to simulate normal operation. Between these ballast and de-ballast operations, the vessel would conduct voyages between at least two different ports as described in Appendix A of the QAPP (Appendix C in the present report). The

CB75/250 combination was installed in two 20' containers placed in the aft part of the lower hold and connected to the ballast water system of the vessel. For the shipboard testing, the ballast tank pairs 7 and 8 were used for treated water and the ballast tank pair 3 was used for control water. The individual tests with the CB250 were conducted as presented in Table 5.1.

Table 5.1 Details of inlet and discharge operations for shipboard test cycles

Test cycle	Location	Operation	Inlet	Volume and flow rate	Discharge	Volume and flow rate
CB250-Test#1	Hundested	Control	2011.10.28 12:08-13:13	268 m^3 247 m^3/h	2011.10.29 14:07-14:57	210 m^3 252 m^3/h
		Treatment	2011.10.28 13:25-15:43	582 m^3 253 m^3/h	2011.10.29 11:50-13:54	550 m^3 266 m^3/h
CB250-Test#2	Hundested	Control	2011.10.31 10:03-11:08	270 m^3 249 m^3/h	2011.11.01 11:10-12:10	250 m^3 250 m^3/h
		Treatment	2011.10.31 11:21-13:45	600 m^3 250 m^3/h	2011.11.01 08:38-10:49	524 m^3 240 m^3/h
CB250-Test#3	Hundested	Control	2011.11.02 09:59-11:04	270 m^3 249 m^3/h	2011.11.03 10:49-11:49	250 m^3 250 m^3/h
		Treatment	2011.11.02 11:26-13:26	500 m^3 250 m^3/h	2011.11.03 08:37-10:24	445 m^3 250 m^3/h
CB250-Test#4	Hundested	Control	2011.11.10 09:52-10:52	250 m^3 250 m^3/h	2011.11.11 10:42-11:34	217 m^3 250 m^3/h
		Treatment	2011.11.10 11:14-12:59	440 m^3 251 m^3/h	2011.11.11 08:58-10:29	376 m^3 248 m^3/h
CB250-Test#5	Hundested	Control	2012.04.28 08:40-09:18	180 m^3 284 m^3/h	2012.04.29 09:48-10:21	130 m^3 230 m^3/h
		Treatment	2012.04.28 09:36-11:08	384 m^3 250 m^3/h	2012.04.29 08:25-09:35	307 m^3 263 m^3/h
CB250-Test#6	Hundested	Control	2012.04.30 09:37-10:15	180 m^3 284 m^3/h	2012.05.01 09:57-10:27	135 m^3 270 m^3/h
		Treatment	2012.04.30 10:37-12:08	380 m^3 251 m^3/h	2012.05.01 08:34-09:45	291 m^3 246 m^3/h

Ballast and de-ballast operations were conducted while the vessel was docked in the port of Hundested, Denmark. The holding time varied from 19 to 21 hours for treated water and from 23 to 25 hours for control water. Each test cycle consisted of sampling and analyses of:

- **Inlet water**: Physical-chemical and biological parameters in the inlet water were considered sufficiently stable during the ballasting and, thus, only one set of samples and analyses was used to represent the control tank and the ballast tank
- **Control discharge water**: Stored without treatment from the time of ballasting to discharge
- **Treated discharge water**: Treated and stored from the time of ballasting to discharge

5.2 Sampling

5.2.1 Sample overview

All samples were collected by DHI staff in accordance with the description in the QAPP (Appendix C).

Table 5.2 Number of samples and sample volumes

Water type	Sample replicates	Sample volume per replicate
Inlet water	3 replicates	Organisms ≥50 µm: >1 m^3 *
		Organisms ≥10 and <50 µm: >1 L **
		Bacteria: >0.5 L **
		DOC + POC: Approx. 0.5 L **
		TSS: Approx. 2 L **
Control discharge water	3 replicates	Organisms ≥50 µm: >1 m^3 *
		Organisms ≥10 and <50 µm: >1 L **
		Bacteria: >0.5 L **
		DOC + POC: Approx. 0.5 L **
		TSS: approx. 2 L **
Treated discharge water	3 replicates	Organisms ≥50 µm: >3 m^3 *
	3 × 3 replicates	Organisms ≥10 and <50 µm: >1 L **
	3 × 3 replicates	Bacteria: >0.5 L **
	3 replicates	DOC + POC: Approx. 0.5 L **
	3 replicates	TSS: Approx. 2 L **

* Samples collected by continuous flow during the entire period of intake or discharge; this continuous sampling of 3 replicates, each with a volume >3 m^3, provides the same statistical basis for counting as the sampling 3 × 3 replicates of >1 m^3, which is recommended in the IMO G8 guidelines
** Samples collected over the period of intake or discharge (start, middle and end)
DOC Dissolved organic carbon
POC Particulate organic carbon
TSS Total suspended solids

5.2.2 Samples for enumeration of organisms ≥50 µm

Three replicates were collected by parallel continuous sampling during the entire periods of inlet and discharge. The samples were gently filtered through a net with a mesh size of 35 µm and a reservoir (cod-end) at the bottom for collecting the organisms ≥50 µm. Each replicate was transferred to 1-L glass bottles. The total volume of the filtered sample exceeded 3 m^3 per replicate for the treated discharge samples and 1 m^3 per replicate for the inlet and control discharge samples. The exact sample volume for each of the three replicates was determined by use of three flow meters, which were connected to the relevant sampling ports installed for the systems.

5.2.3 Samples for enumeration of organisms ≥10 µm and <50 µm

Samples (3 replicates for the inlet water, 3 replicates for the control discharge water, and 3 × 3 replicates for the treated discharge water) with a volume of approx. 10 L were collected in polyethylene containers.

5.2.4 Samples for enumeration of bacteria

Samples (3 replicates for each water type) with a volume of at least 0.5 L were collected in sterile polyethylene containers.

5.2.5 Samples for DOC, POC and TSS analyses

Samples (3 replicates for the inlet water, 3 replicates for the control discharge water, and 3 replicates for the treated discharge water) were collected in heat-sterilized blue cap bottles of at least 0.5 L for analysis of DOC and POC. For TSS analysis, samples with a volume of approx. 2 L were collected in polyethylene containers.

5.3 Analyses

5.3.1 Analysis overview

Table 5.3 Overview of analyses and sample replicates

Replicate	Temperature	Salinity	≥ 50 µm	10-50 µm primary production	10-50 µm, MPN	10-50 µm, Lugol's	Bacteria	DOC + POC	TSS
Inlet water									
Replicate 1 (start)	1	1	Three repli-cates	1	1	1	1	1	1
Replicate 2 (mid)	2	2		2	2	2	2	2	2
Replicate 3 (end)	3	3		3	3	3	3	3	3
Control discharge water									
Replicate 1 (start)	1	1	Three repli-cates	1	1	1	1	1	1
Replicate 2 (mid)	2	2		2	2	2	2	2	2
Replicate 3 (end)	3	3		3	3	3	3	3	3
Treated discharge water									
Replicate 1-3 (start)	1	1	Three repli-cates	1	1-3	-	1-3	1	1
Replicate 4-6 (mid)	4	4		4	4-6	-	4-6	4	4
Replicate 7-9 (end)	7	7		7	7-9	-	7-9	7	7

MPN Most probable number
DOC Dissolved organic carbon
POC Particulate organic carbon
TSS Total suspended solids

All analyses were carried out in accordance with the QAPP (Appendix C) and the relevant standard operating procedures (SOPs). Samples were transported from Hundested to the DHI laboratory in Hørsholm where the analyses were performed.

5.3.2 Organism size class ≥50 µm

The concentrations of viable organisms ≥50 µm in the samples were determined by using a stereo microscope and a counting chamber. Viable organisms were determined after staining with Neutral Red on the basis of observed mobility and morphology according to SOP 30/1700. The viable organisms were characterized according to broad taxonomic groups. Compliance with the IMO D-2 standard (IMO 2004) was verified by using the direct count of viable organisms ≥50 µm.

5.3.3 Organism size class ≥10 µm and <50 µm

The polyethylene container with an approximate sample volume of 10 L was gently turned upside down five times, after which subsamples were taken for the analyses described below.

Organisms in inlet water

One subsample per replicate, with a volume of approx. 10 mL, was transferred to 10-mL polyethylene tubes with screw-caps. The concentrations of viable algae were determined by use of a most probable number (MPN) assay. A dilution series was made for each replicate and 1-mL aliquots containing 1 mL, 0.1 mL and 0.01 mL of the subsample were added to series of five test tubes with 5 mL of liquid medium. Blank controls containing 5 mL of liquid medium without sample were also prepared. The fluorescence of the test tubes was determined prior to the incubation. The concentrations of viable algae in the samples were determined by measuring the fluorescence in the test tubes after 14 days of incubation under continuous light. The incubation of the test tubes was conducted at 13-15°C.

The concentrations of organisms in the size class ≥10 and <50 µm were determined by inverted microscopy. Subsamples with a volume of approx. 100 mL were transferred from the inlet water samples to brown 100-mL glass bottles. The subsamples were preserved by addition of Lugol's solution to achieve 2% final concentration. The identification comprised detailed examination of the algal chloroplasts to confirm that the organisms had been alive and classification of the algae in major taxonomic groups.

The primary production was determined by measuring the ^{14}C fixed by photosynthesis. Two subsamples per replicate, with a volume of approx. 60 mL, were transferred to 60-mL bottles and $NaH^{14}CO_3$ (2 µCi) was added to each bottle and the bottles were incubated for approx. 75 min under light from a light-panel. During the testing in October and November 2011, the incubation was conducted at 9-11°C. During the shipboard testing in April and May 2012, the incubation was conducted at 9-12°C. After incubation, the samples were filtered onto Whatman GF/D filters, and the filters were transferred to glass vials, after which 200 µL 0.1 N HCl was added directly to the filters. The ^{14}C activity remaining in the algae on the filters after acidification was quantified by liquid scintillation counting.

Organisms in discharge water
The concentrations of viable algae in the control discharge water and the treated discharge water were determined by use of the MPN assay. Compliance with the IMO D-2 standard (IMO 2004) was verified by using the total of viable algae determined by the MPN assay and viable micro-zooplankton ≥10 and <50 µm as described in SOPs 30/1701 and 30/1704. As a supporting parameter, measurements of primary production were conducted by use of the method described above.

Furthermore, organisms in the size class ≥10 and <50 µm in the control discharge water samples were also counted by inverted microscopy after fixation with Lugol's solution.

5.3.4 Bacteria

The concentrations of *E. coli* and enterococci were determined by diluting the samples in sterilized water (1:1), after which the samples were distributed in a specific 96-wells test kit for either *E. coli* or enterococci (BIO-RAD, MUG/MUD kits for *E. coli* or enterococci quantification). The inoculated test kits were incubated for 36 hours at 44°C (42°C to 44.5°C), and then positive wells were used to calculate the most probable numbers.

Samples for detection of *Vibrio cholerae* were filtered through a 0.45-µm filter, after which the filters were submerged into alkaline saline peptone water for two selective enrichments. The cultures obtained by the enrichments were used for inoculation of agar plates. Following 24 hours of incubation at 37 ± 1°C, the morphology of the colonies on the agar plates was inspected.

5.3.5 Physical-chemical parameters

Dissolved oxygen, salinity, temperature and pH were measured by use of portable multi parameter instrument equipped with electrodes. In the test cycles #5 and #6 (see Table 5.1), turbidity was also measured. Measurements were conducted at regular intervals throughout the inlet and discharge operations.

Samples for determination of organic carbon content were filtered through a 0.45-µm syringe filter. By using a TOC analyser, the TOC was determined by analysis of non-filtered sample whereas the dissolved organic carbon (DOC) was determined by analysis of filtered sample. The particulate organic carbon (POC) was calculated as the difference between TOC and DOC.

Samples for determination of TSS were filtered through a glass fibre filter, which had already been weighed in the laboratory, and the TSS was determined by weighing of filters containing sample after drying at 105°C.

6 Results

6.1 Physical-chemical parameters

The physical-chemical conditions of inlet and discharge waters for test cycles with the CB250 are summarized in Table 6.1 and Table 6.2. Onsite measurement data are also available in the data logging in Appendix A. Detailed data for TSS, POC and DOC are available in Appendix B.

Table 6.1 Average concentrations of total suspended solids (TSS), particulate organic carbon (POC) and dissolved organic carbon (DOC).

Test cycle	Water type	TSS (mg/L)	POC (mg/L)	DOC (mg/L)
CB250-Test#1	Inlet water	1.1	0.57	1.8
	Control discharge water	1.5	0.55	1.8
	Treated discharge water	1.6	0.51	2.6
CB250-Test#2	Inlet water	1.1	0.25	3.0
	Control discharge water	1.6	0.27	2.9
	Treated discharge water	1.2	0.34	2.7
CB250-Test#3	Inlet water	0.91	0.31	3.3
	Control discharge water	1.1	0.21	3.0
	Treated discharge water	1.2	0.15	3.0
CB250-Test#4	Inlet water	1.0	0.23	2.2
	Control discharge water	1.1	0.23	2.2
	Treated discharge water	1.3	0.20	2.3
CB250-Test#5	Inlet water	1.7	0.42	3.1
	Control discharge water	0.77	0.44	2.9
	Treated discharge water	1.4	0.29	3.2
CB250-Test#6	Inlet water	2.0	0.39	3.1
	Control discharge water	1.5	0.27	3.1
	Treated discharge water	1.5	0.30	3.4

Table 6.2 Average measurements of oxygen (O_2), salinity, temperature, pH and UV transmittance (UVT)

Test cycle	Water type	O_2 (%)	Salinity (PSU)	Temper-ature (°C)	pH	UVT (%)	Turbidity (NTU)
CB250-Test#1	Inlet water	86	25.8	11.6	7.9	91	-
	Control discharge water	84	25.7	11.2	7.9	89	-
	Treated discharge water	89	22.2	11.1	7.9	89	-
CB250-Test#2	Inlet water	95	20.3	10.0	8.0	89	-
	Control discharge water	96	20.4	10.2	8.0	89	-
	Treated discharge water	95	20.6	10.6	8.0	89	-
CB250-Test#3	Inlet water	96	19.0	9.9	8.0	86	-
	Control discharge water	96	19.0	10.1	8.0	86	-
	Treated discharge water	95	19.2	10.3	8.0	86	-
CB250-Test#4	Inlet water	90	22.5	10.4	-	88	-
	Control discharge water	90	22.5	10.1	-	90	-
	Treated discharge water	89	22.3	10.1	-	91	-
CB250-Test#5	Inlet water	98	16.1	9.1	8.2	88	1
	Control discharge water	97	15.3	9.3	8.2	88	2
	Treated discharge water	98	16.7	9.5	8.2	91	1
CB250-Test#6	Inlet water	98	17.2	9.4	-	89	1
	Control discharge water	94	16.7	9.6	8.2	89	2
	Treated discharge water	98	17.7	10.3	8.2	89	1

PSU Practical salinity units
NTU Nephelometric turbidity units

6.2 Biological parameters

The densities of live organisms in the inlet and control discharge water were in accordance with the IMO G8 guidelines in all test cycles. Detailed data from the biological efficacy analyses are available in Appendix B.

6.2.1 Organism size class ≥50 µm

The average densities of viable organisms in the ≥50 µm size class varied from approx. 3,500 to 29,000 organisms per m^3 in the inlet water and from approx. 1,900 to 22,000 organisms per m^3 in the control discharge water (Table 6.3). The majority of organisms ≥50 µm in the inlet water were identified as copepods, nauplii, and polychaete larvae. In test cycles #4, #5 and #6, rotifers also contributed significantly to the total number of organisms ≥50 µm.

Table 6.3 Total sample volumes and average concentrations of viable organisms in the size class ≥50 µm. Specific data and individual sample volumes are provided in Appendix B.

Test cycle	Inlet water		Control discharge water		Treated discharge water	
	Total sample volume (m^3)	Organ-isms/m^3	Total sample volume (m^3)	Organ-isms/m^3	Total sample volume (m^3)	Organ-isms/m^3
CB250-Test#1	3.718	10,256	3.547	5,553	10.092	0.20
CB250-Test#2	3.763	3,518	3.873	1,897	10.311	0
CB250-Test#3	3.711	14,115	3.617	11,466	9.573	0
CB250-Test#4	3.523	7,811	3.593	6,368	9.627	0.21
CB250-Test#5	3.327	29,037	3.696	21,517	10.023	0
CB250-Test#6	3.515	21,651	3.323	12,676	9.873	1.9
IMO G8	>3	>90	>3	>10	>9	<10

In test cycles #2, #3 and #5, no viable organisms ≥50 µm were identified in the treated discharge samples. In test cycles #1, #4 and #6, the average numbers of viable organisms in the ≥50 µm size class were 0.20, 0.21 and 1.9 per m^3, respectively, which is 5-50 times below the threshold value defined in the IMO D-2 standard.

6.2.2 Organism size class ≥10 and <50 µm

For the size class ≥10 and <50 µm, the average densities in inlet water varied from approx. 110 to 400 organisms/mL when determined by inverted microscopy, and from 140 to >160 organisms/mL when determined by the MPN assay (Table 6.4). The average densities of viable algae in the control discharge water determined by MPN varied from 102 to >160 organisms/mL. Furthermore, inverted microscopy showed densities from approx. 50 to 360 organisms/mL in the control discharge water samples.

The MPN of algae in the treated discharge water was below the MPN assay detection limit of 0.18 organisms/mL in four out of six test cycles. In test cycle #4 and #6 the average densities of viable algae determined by MPN were 0.48 and 0.21 organisms/mL, respectively. The MPN of the individual replicates in test cycle #4 were 1.3 organisms/mL in two replicates, 0.40 organisms/mL in two replicates, and <0.18 organisms/mL in five replicates, and the average MPN was expressed as 0.48 organisms/mL by using the value 0.18 organisms/mL for the replicates showing values below the detection limit. The MPN of the individual replicates in test cycle #6 were 0.45 organisms/mL in one replicate and <0.18 in the other eight replicates (Appendix B). Micro-zooplankton ≥10 and <50 µm was not observed in the treated discharge water. By using the results of the MPN assay, the average densities of viable organisms representing the ≥10 and <50 µm size class in the treated discharge water were thus 20-55 times below the IMO D-2 standard.

Table 6.4 Average concentrations of viable organisms in the size class ≥10 µm and <50 µm. The primary production decrease is expressed as the percentage reduction of the measured primary production in inlet water. Specific data are provided in Appendix B.

Test cycle	Water type	Microscopy (organ-isms/mL)	MPN (organ-isms/mL)	Primary production	
				DPM	Decrease (%)
CB250-Test#1	Inlet water	134	>160	487	100
	Control discharge water	98	>160	288	
	Treated discharge water	-	<0.18	0.21	
CB250-Test#2	Inlet water	209	>160	666	99.3
	Control discharge water	161	>160	530	
	Treated discharge water	-	<0.18	4.5	
CB250-Test#3	Inlet water	212	>160	1,012	99.6
	Control discharge water	291	>160	981	
	Treated discharge water	-	<0.18	4.4	
CB250-Test#4	Inlet water	108	140	266	98.7
	Control discharge water	46	>160	376	
	Treated discharge water	-	0.48	3.4	
CB250-Test#5	Inlet water	117	>160	808	100
	Control discharge water	91	>160	607	
	Treated discharge water	-	<0.18	0	
CB250-Test#6	Inlet water	403	>160	637	99.7
	Control discharge water	356	102	532	
	Treated discharge water	-	0.21	2.0	
IMO G8	Inlet water	>90	-	-	-
	Control discharge water	-	>10	-	-
	Treated discharge water	-	<10	-	-

MPN Most probable number
DPM Disintegrations per minute

Table 6.5 Algal species identified in inlet water and their ability to grow under the conditions applied in the MPN assay

Groups and species	CB250-Test#1	CB250-Test#2	CB250-Test#3	CB250-Test#4	CB250-Test#5	CB250-Test#6	Capable of growing in MPN assay
Bacillariophyceae							
Ørataulina pelagica				X			+
Chaetoceros sp.	X					X	
Chaetoceros wighamii		X					+
Chaetoceros, solitary species			X				
Leptocylindrus minimus		X		X			+
Navicula spp.				X			+
Phaeodactylum tricornutum			X				+
Pseudonitzschia sp.		X					+
Rhizosolenia setigera			X				
Rhizosolenia styliformis	X						
Skeletonema costatum	X	X	X		X	X	+
Thalassiosira spp.	X		X				+
Dictyochophyceae							
Dictyocha speculum		X	X				
Dinophyceae							
Ceratium furca	X	X					+
Heterocapsa triquetra			X				+
Katodinium glaucum		X					+
Katodinium sp.						X	+
Prorocetrum micans			X				+
Cryptophyceae							
Cryptophytes spp.	X		X	X	X	X	+

Growth under the conditions applied in the MPN assay was confirmed for 74% of the algal species identified in the inlet water (Table 6.5). The decrease in the primary production of approx. 99-100% during treatment in the BWMS confirmed that the treatment resulted in an immediate impact on the algal photosynthesis.

UV irradiation causes damage of the DNA in the cells and it may take several days before the cell membrane is disrupted and the enzyme activity stops (Stehouwer et al. 2010, Liltved et al. 2011, Liebich et al. 2012). Enumeration of algae by use of the MPN assay is directly related to growth over a certain time period. The ability of algal species to grow is a meaningful definition of viability in an evaluation, of which the target is to determine the efficiency of treatment aiming to reduce the species in ballast water capable to proliferate and survive in the natural environment. For UV treatment systems, the MPN assay is considered the best available methodology for evaluation of viable algae. Primary production analyses provide a measure for algal photosynthesis by determining amounts of ^{14}C fixed by photosynthesis. Neither the MPN assay nor primary production analyses are limited to the ≥10 and <50 µm size class; on the contrary, these parameters include planktonic algae without reference to size.

6.2.3 Bacteria

For shipboard testing, there are no requirements in the IMO G8 guidelines in relation to the density of bacteria in the inlet water or the control discharge water.

Table 6.6 Average bacterial concentrations. Specific data are provided in Appendix B.

Test cycle	Water type	*E. coli* (CFU/100 mL)	Enterococci (CFU/100 mL)	*Vibrio cholerae* (CFU/100 mL)
CB250-Test#1	Inlet water	17	<10	<1
	Control discharge water	14	10	<1
	Treated discharge water	<10	<10	<1
CB250-Test#2	Inlet water	<10	<10	<1
	Control discharge water	43	<10	<1
	Treated discharge water	<10	<10	<1
CB250-Test#3	Inlet water	54	<10	<1
	Control discharge water	47	<10	<1
	Treated discharge water	<10	<10	<1
CB250-Test#4	Inlet water	10	<10	<1
	Control discharge water	<10	<10	<1
	Treated discharge water	<10	<10	<1
CB250-Test#5	Inlet water	14	<10	<1
	Control discharge water	<10	<10	<1
	Treated discharge water	<10	<10	<1
CB250-Test#6	Inlet water	14	<10	<1
	Control discharge water	<10	10	<1
	Treated discharge water	<10	<10	<1
IMO G8	Inlet water	-	-	-
	Control discharge water	-	-	-
	Treated discharge water	<250	<100	<1

CFU Colony-forming units

The contents of *E. coli* in the inlet water and control discharge water were generally low and enterococci were hardly detected. In the treated discharge water the average contents of *E. coli* and enterococci were below the detection limit in all test cycles. *Vibrio cholerae* was not identified in any of the test cycles.

7 Conclusion

The ability of the CB250 to (a) successfully treat ballast water without interruption and (b) meet IMO D-2 ballast water discharge standard was evaluated during a series of six consecutive valid test cycles.

The average densities of viable organisms in the ≥50 µm size class varied from approx. 3,500 to approx. 29,000 organisms per m^3 in the inlet water. For the size class ≥10 and <50 µm, the average densities in inlet water varied from approx. 110 to 400 organisms/mL when determined by inverted microscopy. The contents of *E. coli* in the inlet water were generally low and enterococci were hardly detected. The inlet water concentrations of organisms ≥50 µm and the smaller planktonic organisms (≥10 and <50 µm) fulfilled the validity criteria defined in the IMO G8 guidelines.

The numbers of viable organisms in the ≥50 μm size class were 0 per m^3 in three test cycles and 0.20-1.9 per m^3 in the three remaining test cycles, which is 5-50 times below the threshold value defined in the IMO D-2 standard. The density of viable algae in the treated discharge water was determined to <0.18-0.48 organisms/mL by use of a most probable number (MPN) assay, and micro-zooplankton ≥10 and <50 μm was not observed in the treated discharge water. By using the results of the MPN assay, the density of viable organisms representing the ≥10 and <50 μm size class in the treated discharge water was thus 20-55 times below the IMO D-2 standard. Measurements of primary production showed a decrease of 99-100% after treatment in the BWMS compared with the control with untreated ballast water, which confirmed that the treatment resulted in an immediate impact on the algal photosynthesis. In the treated discharge water the average contents of *E. coli* and enterococci were below the detection limit in all test cycles. *Vibrio cholerae* was not identified in any of the test cycles.

The CB250 functioned properly during the six test cycles and was highly effective at reducing live organism densities under the shipboard testing conditions. The densities of live organisms in the size classes and the densities of specific bacteria defined in the IMO G8 guidelines were below the IMO D-2 standard in the treated discharge water in all test cycles. All six test cycles are considered to fulfil the validity criteria of the IMO G8 guidelines.

8 References

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A P P E N D I X   A

Data logging from the shipboard testing with CrystalBallast®250

Table A.1.1 Test cycle data logging; treated water

Subject	Results		Comments
Client treatment system	Auramarine CB250		Filter: FilterSafe BSF-100H; 40µm
Salinity (PSU)	26		
Ballast tank No.	Pairs 7 & 8		
Test cycle No.	#1		
Location for intake	Hundested		
Date and time intake start	2011.10.28	13:25	
Date and time intake stop	2011.10.28	15.43	
Flow during intake	253 m^3/h		
Treated volume during intake	582 m^3		
Location for discharge	Hundested		
Date and time discharge start	2011.10.29	11:50	
Date and time discharge stop	2011.10.29	13:54	
Flow during discharge	266 m^3/h		
Treated volume during discharge	550 m^3		

Table A.1.2 Test cycle data logging; control water

Subject	Results		Comments
Salinity (PSU)	22		
Ballast tank No.	Pair 3		
Date and time intake start	2011.10.28	12:08	
Date and time intake stop	2011.10.28	13.13	
Flow during intake	247 m^3/h		
Volume during intake	268 m^3		
Date and time discharge start	2011.10.29	14:07	
Date and time discharge stop	2011.10.29	14:57	
Flow during discharge	252 m^3/h		
Volume during discharge	210 m^3		

Table A.1.3 Test cycle data logging; onsite measurements

Water type	Dissolved oxygen (%)	Salinity (PSU)	Temperature (°C)	pH	UVT (%)	UV intensity (mW/cm^2)
Inlet water (control)	86 (0.58)	25.8 (0.25)	11.6 (0.16)	7.9 (0.05)	91 (-)	-
Inlet water (treated)	90 (1.3)	23.8 (1.1)	11.1 (0.25)	8.0 (0.05)	88 (-)	107 (20)
Control discharge	84 (0.58)	25.7 (-)	11.2 (0.15)	7.9 (-)	89 (-)	-
Treated discharge	89 (0.41)	22.2 (0.19)	11.1 (0.16)	7.9 (0.14)	89 (-)	92 (3.9)

( ) Standard deviation
PSU Practical salinity units
UVT UV transmittance

Table A.2.1 Test cycle data logging; treated water

Subject	Results	Comments
Client treatment system	Auramarine CB250	Filter: Boll&Kirch 6.18.2; 30 µm
Salinity (PSU)	20	
Ballast tank No.	Pairs 7 & 8	
Test cycle No.	#2	
Location for intake	Hundested	
Date and time intake start	2011.10.31 11:21	
Date and time intake stop	2011.10.31 13.45	
Flow during intake	250 m^3/h	
Treated volume during intake	600 m^3	
Location for discharge	Hundested	
Date and time discharge start	2011.11.01 08:38	
Date and time discharge stop	2011.11.01 10:49	
Flow during discharge	240 m^3/h	
Treated volume during discharge	524 m^3	

Table A.2.2 Test cycle data logging; control water

Subject	Results	Comments
Salinity (PSU)	20	
Ballast tank No.	Pair 3	
Date and time intake start	2011.10.31 10:03	
Date and time intake stop	2011.10.31 11.08	
Flow during intake	249 m^3/h	
Volume during intake	270 m^3	
Date and time discharge start	2011.11.01 11:10	
Date and time discharge stop	2011.11.01 12:10	
Flow during discharge	250 m^3/h	
Volume during discharge	250 m^3	

Table A.2.3 Test cycle data logging; onsite measurements

Water type	Dissolved oxygen (%)	Salinity (PSU)	Tempera-ture (°C)	pH	UVT (%)	UV intensity (mW/cm^2)
Inlet water (control)	95 (0.55)	20.3 (0.07)	10.0 (0.04)	8.0 (0.04)	89 (-)	-
Inlet water (treated)	95 (1.0)	20.4 (0.06)	10.2 (0.06)	8.0 (-)	90 (-)	109 (2.1)
Control discharge	96 (0.58)	20.4 (0.06)	10.2 (-)	8.0 (-)	89 (-)	-
Treated discharge	95 (0.55)	20.6 (-)	10.6 (-)	8.0 (-)	89 (-)	97 (10)

( ) Standard deviation
PSU Practical salinity units
UVT UV transmittance

Table A.3.1 Test cycle data logging; treated water

Subject	Results		Comments
Client treatment system	Auramarine CB250		Filter: Boll&Kirch 6.18.2; 30 µm
Salinity (PSU)	19		
Ballast tank No.	Pairs 7 & 8		
Test cycle No.	#3		
Location for intake	Hundested		
Date and time intake start	2011.11.02	11:26	
Date and time intake stop	2011.11.02	13.26	
Flow during intake	250 m³/h		
Treated volume during intake	500 m³		
Location for discharge	Hundested		
Date and time discharge start	2011.11.03	08:37	
Date and time discharge stop	2011.11.03	10:24	
Flow during discharge	250 m³/h		
Treated volume during discharge	445 m³		

Table A.3.2 Test cycle data logging; control water

Subject	Results		Comments
Salinity (PSU)	19		
Ballast tank No.	Pair 3		
Date and time intake start	2011.11.02	09:59	
Date and time intake stop	2011.11.02	11.04	
Flow during intake	249 m³/h		
Volume during intake	270 m³		
Date and time discharge start	2011.11.03	10:49	
Date and time discharge stop	2011.11.03	11:49	
Flow during discharge	250 m³/h		
Volume during discharge	250 m³		

Table A.3.3 Test cycle data logging; onsite measurements

Water type	Dissolved oxygen (%)	Salinity (PSU)	Tempera-ture (°C)	pH	UVT (%)	UV intensity (mW/cm²)
Inlet water (control)	96 (0.45)	19.0 (0.08)	9.9 (0.05)	8.0 (0.04)	86 (-)	-
Inlet water (treated)	96 (-)	19.1 (0.06)	10.1 (-)	8.0 (-)	87 (-)	88 (0.58)
Control discharge	96 (0.71)	19.0 (-)	10.1 (-)	8.0 (-)	86 (-)	-
Treated discharge	95 (0.71)	19.2 (-)	10.3 (0.04)	8.0 (0.04)	86 (-)	78 (8.4)

( ) Standard deviation
PSU Practical salinity units
UVT UV transmittance

Table A.4.1 Test cycle data logging; treated water

Subject	Results		Comments
Client treatment system	Auramarine CB250		Filter: FilterSafe BSF-100H; 40µm
Salinity (PSU)	20		
Ballast tank No.	Pairs 7 & 8		
Test cycle No.	#4		
Location for intake	Hundested		
Date and time intake start	2011.11.10	11:14	
Date and time intake stop	2011.11.10	12.59	
Flow during intake	251 m³/h		
Treated volume during intake	440 m³		
Location for discharge	Hundested		
Date and time discharge start	2011.11.11	08:58	
Date and time discharge stop	2011.11.11	10:29	
Flow during discharge	248 m³/h		
Treated volume during discharge	376 m³		

Table A.4.2 Test cycle data logging; control water

Subject	Results		Comments
Salinity (PSU)	20		
Ballast tank No.	Pair 3		
Date and time intake start	2011.11.10	09:52	
Date and time intake stop	2011.11.10	10.52	
Flow during intake	250 m³/h		
Volume during intake	250 m³		
Date and time discharge start	2011.11.11	10:42	
Date and time discharge stop	2011.11.11	11:34	
Flow during discharge	250 m³/h		
Volume during discharge	217 m³		

Table A.4.3 Test cycle data logging; onsite measurements

Water type	Dissolved oxygen (%)	Salinity (PSU)	Tempera-ture (°C)	pH	UVT (%)	UV intensity (mW/cm²)
Inlet water (control)	90 (1.0)	22.5 (0.05)	10.4 (-)	-	88 (-)	-
Inlet water (treated)	89 (0.58)	22.4 (0.06)	10.3 (0.29)	-	87 (-)	118 (4.0)
Control discharge	90 (0.58)	22.5 (-)	10.1 (0.06)	-	90 (-)	-
Treated discharge	89 (0.58)	22.3 (-)	10.1 (0.05)	-	91 (-)	112 (1.7)

( ) Standard deviation
PSU Practical salinity units
UVT UV transmittance

Table A.5.1 Test cycle data logging; treated water

Subject	Results		Comments
Client treatment system	Auramarine CB250		Filter: FilterSafe BSF-100H; 40µm
Salinity (PSU)	17		
Ballast tank No.	Pair 7 & 8		
Test cycle No.	#5		
Location for intake	Hundested		
Date and time intake start	2012.04.28	09:36	
Date and time intake stop	2012.04.28	11:08	
Flow during intake	250 m^3/h		
Treated volume during intake	384 m^3		
Location for discharge	Hundested		
Date and time discharge start	2012.04.29	08:25	
Date and time discharge stop	2012.04.29	09:35	
Flow during discharge	263 m^3/h		
Treated volume during discharge	307 m^3		

Table A.5.2 Test cycle data logging; control water

Subject	Results		Comments
Salinity (PSU)	15		
Ballast tank No.	Pair 3		
Date and time intake start	2012.04.28	08:40	
Date and time intake stop	2012.04.28	09:18	
Flow during intake	284 m^3/h		
Volume during intake	180 m^3		
Date and time discharge start	2012.04.29	09:47	
Date and time discharge stop	2012.04.29	10:21	
Flow during discharge	230 m^3/h		
Volume during discharge	130 m^3		

Table A.5.3 Test cycle data logging; onsite measurements

Water type	Dissolved oxygen (%)	Salinity (PSU)	Tempera-ture (°C)	pH	Turbidity (NTU)	UVT (%)	UV intensity (W/m^2)
Inlet water (control)	97 (±1.0)	15.4 (±0.70)	9.0 (±0.15)	8.2 (±0.06)	1 (-)	88 (-)	-
Inlet water (treated)	98 (-)	16.7 (±0.35)	9.1 (-)	8.2 (-)	1 (-)	-	146 (±1.7)
Control discharge	97 (±1.0)	15.3 (-)	9.3 (±0.06)	8.2 (-)	2 (-)	88 (-)	-
Treated discharge	98 (-)	16.7 (-)	9.5 (±0.10)	8.2 (±0.06)	1 (-)	91 (-)	137 (±1.2)

( ) Standard deviation
PSU Practical salinity units
NTU Nephelometric turbidity units
UVT UV transmittance

Table A-6.1 Test cycle data logging; treated water

Subject	Results		Comments
Client treatment system	Auramarine CB250		Filter: Boll&Kirch 6.18.2; 30 µm
Salinity (PSU)	18		
Ballast tank No.	Pair 7 & 8		
Test cycle No.	#6		
Location for intake	Hundested		
Date and time intake start	2012.04.30	10:37	
Date and time intake stop	2012.04.30	12:08	
Flow during intake	251 m^3/h		
Treated volume during intake	380 m^3		
Location for discharge	Hundested		
Date and time discharge start	2012.05.01	08:34	
Date and time discharge stop	2012.05.01	09:45	
Flow during discharge	246 m^3/h		
Treated volume during discharge	291 m^3		

Table A.6.2 Test cycle data logging; control water

Subject	Results		Comments
Salinity (PSU)	17		
Ballast tank No.	Pair 3		
Date and time intake start	2012.04.30	09:37	
Date and time intake stop	2012.04.30	10:15	
Flow during intake	284 m^3/h		
Volume during intake	180 m^3		
Date and time discharge start	2012.05.01	09:57	
Date and time discharge stop	2012.05.01	10:27	
Flow during discharge	270 m^3/h		
Volume during discharge	135 m^3		

Table A.6.3 Test cycle data logging; onsite measurements

Water type	Dissolved oxygen (%)	Salinity (PSU)	Temperature (°C)	pH	Turbidity (NTU)	UVT (%)	UV intensity (W/m^2)
Inlet water (control)	97 (±1.2)	16.6 (±0.36)	9.2 (±0.06)	-	1 (-)	89 (-)	-
Inlet water (treated)	98 (±0.58)	17.8 (±0.06)	9.6 (±0.10)	-	1 (-)	-	133 (±1.5)
Control discharge	94 (-)	16.7 (±0.06)	9.6 (±0.06)	8.2 (-)	2 (-)	89 (-)	-
Treated discharge	98 (-)	17.7 (-)	10.3 (±0.12)	8.2 (-)	1 (-)	89 (-)	129 (±0.58)

( ) Standard deviation
PSU Practical salinity units
NTU Nephelometric turbidity units
UVT UV transmittance

APPENDIX B

Detailed data for physical and chemical properties and biological efficacy analyses in shipboard testing of CrystalBallast®250

Physical-chemical parameters

Table B.1 Measurements of total suspended solids (TSS)

Test cycle	Water type	TSS (mg/L)				
		FR1	FR2	FR3	AVG	STD
#1	Inlet water	0.91	1.4	1.1	**1.1**	±0.25
	Control discharge water	1.1	1.3	2.0	**1.5**	±0.44
	Treated discharge water	1.5	1.9	1.6	**1.6**	±0.18
#2	Inlet water	1.4	1.2	0.90	**1.1**	±0.23
	Control discharge water	1.4	1.9	1.5	**1.6**	±0.24
	Treated discharge water	1.5	1.1	1.2	**1.2**	±0.24
#3	Inlet water	1.0	0.85	0.86	**0.91**	±0.10
	Control discharge water	0.92	0.89	1.4	**1.1**	±0.29
	Treated discharge water	1.3	0.87	1.6	**1.2**	±0.34
#4	Inlet water	1.5	1.3	0.25	**1.0**	±0.65
	Control discharge water	1.1	1.2	1.0	**1.1**	±0.13
	Treated discharge water	1.0	1.1	1.7	**1.3**	±0.40
#5	Inlet water	1.8	1.2	2.0	**1.7**	±0.41
	Control discharge water	0.73	0.73	0.83	**0.77**	±0.06
	Treated discharge water	1.2	1.3	1.6	**1.4**	±0.21
#6	Inlet water	2.3	1.9	2.0	**2.0**	±0.20
	Control discharge water	1.6	1.3	1.6	**1.5**	±0.19
	Treated discharge water	1.3	1.6	1.7	**1.5**	±0.19

FR Field replicate
AVG Average
STD Standard deviation

Table B.2 Measurements of particulate organic carbon (POC)

Test cycle	Water type	POC (mg/L)				
		FR1	FR2	FR3	AVG	STD
#1	Inlet water	0.31	0.43	0.96	**0.57**	±0.35
	Control discharge water	0.40	1.1	0.15	**0.55**	±0.49
	Treated discharge water	1.1	0.30	0.13	**0.51**	±0.53
#2	Inlet water	0.15	0.37	0.22	**0.25**	±0.12
	Control discharge water	0.28	0.19	0.33	**0.27**	±0.07
	Treated discharge water	0.39	0.33	0.29	**0.34**	±0.05
#3	Inlet water	0.30	0.28	0.36	**0.31**	±0.04
	Control discharge water	0.24	0.26	0.13	**0.21**	±0.07
	Treated discharge water	0.14	0.09	0.23	**0.15**	±0.07
#4	Inlet water	0.24	0.21	0.24	**0.23**	±0.02
	Control discharge water	0.20	0.29	0.21	**0.23**	±0.05
	Treated discharge water	0.20	0.23	0.17	**0.20**	±0.03
#5	Inlet water	0.34	0.40	0.51	**0.42**	±0.09
	Control discharge water	0.32	0.37	0.63	**0.44**	±0.17
	Treated discharge water	0.16	0.41	0.29	**0.29**	±0.12
#6	Inlet water	0.53	0.31	0.33	**0.39**	±0.12
	Control discharge water	0.19	0.38	0.26	**0.27**	±0.10
	Treated discharge water	0.35	0.31	0.24	**0.30**	±0.06

FR Field replicate
AVG Average
STD Standard deviation

Table B.3 Measurements of dissolved organic carbon (DOC)

Test cycle	Water type	DOC (mg/L)				
		FR1	FR2	FR3	AVG	STD
#1	Inlet water	2.2	1.9	1.5	**1.8**	±0.33
	Control discharge water	2.1	1.2	2.2	**1.8**	±0.51
	Treated discharge water	2.0	2.8	3.1	**2.6**	±0.58
#2	Inlet water	3.2	2.8	2.9	**3.0**	±0.23
	Control discharge water	2.9	2.9	2.8	**2.9**	±0.06
	Treated discharge water	2.7	2.8	2.7	**2.7**	±0.06
#3	Inlet water	3.3	3.4	3.2	**3.3**	±0.10
	Control discharge water	3.0	3.0	3.0	**3.0**	±0.02
	Treated discharge water	3.0	2.9	3.0	**3.0**	±0.03
#4	Inlet water	2.3	2.2	2.2	**2.2**	±0.02
	Control discharge water	2.2	2.1	2.2	**2.2**	±0.05
	Treated discharge water	2.2	2.2	2.3	**2.3**	±0.03
#5	Inlet water	3.5	2.9	2.9	**3.1**	±0.34
	Control discharge water	2.9	2.9	2.8	**2.9**	±0.04
	Treated discharge water	3.7	3.0	2.9	**3.2**	±0.39
#6	Inlet water	3.0	3.1	3.1	**3.1**	±0.07
	Control discharge water	3.2	3.0	3.0	**3.1**	±0.12
	Treated discharge water	3.6	3.1	3.3	**3.4**	±0.23

FR Field replicate
AVG Average
STD Standard deviation

Organism size class ≥50 µm

Table B.4 Enumeration of organisms ≥50 µm and sample volumes

Test cycle	Water type	Organisms ≥50 µm							
		FR1		FR2		FR3		AVG	STD
		vol. m^3	org./m^3	vol. m^3	org./m^3	vol. m^3	org./m^3	org./m^3	
#1	Inlet water	1.234	8,169	1.236	10,340	1.248	12,260	**10,256**	±2,047
	Control discharge water	1.164	5,631	1.180	6,320	1.203	4,708	**5,553**	±809
	Treated discharge water	3.354	0	3.373	0.30	3.365	0.30	**0.20**	±0.17
#2	Inlet water	1.251	4,404	1.255	3,458	1.257	2,692	**3,518**	±858
	Control discharge water	1.293	1,963	1.285	1,417	1.295	2,310	**1,897**	±450
	Treated discharge water	3.434	0	3.444	0	3.433	0	**0**	-
#3	Inlet water	1.234	15,540	1.244	10,710	1.233	16,104	**14,115**	±2,970
	Control discharge water	1.206	12,915	1.207	10,181	1.204	11,302	**11,466**	±1,375
	Treated discharge water	3.189	0	3.196	0	3.188	0	**0**	-
#4	Inlet water	1.171	8,249	1.179	6,521	1.173	8,662	**7,811**	±1,136
	Control discharge water	1.196	6,635	1.200	5,787	1.197	6,683	**6,368**	±504
	Treated discharge water	3.202	0.31	3.221	0	3.204	0.31	**0.21**	±0.18
#5	Inlet water	1.115	27,959	1.101	31,078	1.111	28,073	**29,037**	±1769
	Control discharge water	1.242	23,781	1.243	20,113	1.211	20,657	**21,517**	±1980
	Treated discharge water	3.356	0	3.333	0	3.334	0	**0**	-
#6	Inlet water	1.165	22,721	1.176	21,545	1.174	20,689	**21,651**	±1,020
	Control discharge water	1.093	14,082	1.115	13,399	1.115	10,547	**12,676**	±1,875
	Treated discharge water	3.233	1.5	3.240	1.5	3.400	2.6	**1.9**	±0.64

FR Field replicate
AVG Average
STD Standard deviation

Organism size class ≥10 and <50 μm

Table B.5 Enumeration of organisms ≥10 μm and <50 μm by microscopy

Test cycle	Water type	Organisms/mL				
		FR1	FR2	FR3	AVG	STD
#1	Inlet water	214	87	100	**134**	±70
	Control discharge	153	79	63	**98**	±48
#2	Inlet	237	232	158	**209**	±44
	Control discharge	179	132	174	**161**	±26
#3	Inlet	258	211	168	**212**	±45
	Control discharge	211	295	368	**291**	±79
#4	Inlet	111	105	*	**108**	±3.7
	Control discharge	47	58	32	**46**	±13
#5	Inlet	105	111	134	**117**	±15
	Control discharge	84	89	100	**91**	±8.0
#6	Inlet	387	421	400	**403**	±17
	Control discharge	342	347	379	**356**	±20

FR Field replicate
AVG Average
STD Standard deviation
* Sample lost

Table B.6 Determination of viable algae by the most probable number (MPN) assay

Test cycle	Water type	Viable algae (organisms/mL)									
		FR1	FR2	FR3	FR4	FR5	FR6	FR7	FR8	FR9	AVG
#1	Inlet	>160	>160	>160	-	-	-	-	-	-	**>160**
	Control discharge	>160	>160	>160	-	-	-	-	-	-	**>160**
	Treated discharge	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	**<0.18**
#2	Inlet	>160	>160	>160	-	-	-	-	-	-	**>160**
	Control discharge	>160	>160	>160	-	-	-	-	-	-	**>160**
	Treated discharge	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	**<0.18**
#3	Inlet	>160	>160	>160	-	-	-	-	-	-	**>160**
	Control discharge	>160	>160	>160	-	-	-	-	-	-	**>160**
	Treated discharge	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	**<0.18**
#4	Inlet	160 (54-480)	92 (29-290)	>160	-	-	-	-	-	-	**140**
	Control discharge	>160	>160	>160	-	-	-	-	-	-	**>160**
	Treated discharge	1.3 (0.47-3.5)	0.40 (0.095-1.7)	0.40 (0.095-1.7)	1.3 (0.47-3.5)	<0.18	<0.18	<0.18	<0.18	<0.18	**0.48**
#5	Inlet	>160	>160	>160	-	-	-	-	-	-	**>160**
	Control discharge	>160	160 (54-480)	160 (54-480)	-	-	-	-	-	-	**>160**
	Treated discharge	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	**<0.18**
#6	Inlet	160 (54-480)	160 (54-480)	>160	-	-	-	-	-	-	**>160**
	Control discharge	>160	92 (29-290)	54 (16-180)	-	-	-	-	-	-	**102**
	Treated discharge	0.45 (0.11-1.8)	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	<0.18	**0.21**

FR Field replicate
AVG Average

Table B.7 Measurements of primary production by planktonic algae

Test cycle	Water type	Primary production (DPM)										
		FR1	FR2	FR3	FR4	FR5	FR6	FR7	FR8	FR9	AVG	STD
# 1	Inlet	472	413	574	-	-	-	-	-	-	**487**	±82
	Control discharge	299	248	318	-	-	-	-	-	-	**288**	±36
	Treated discharge	0	-	-	0	-	-	0.63	-	-	**0.21**	±0.36
#2	Inlet	841	616	543	-	-	-	-	-	-	**666**	±155
	Control discharge	455	528	606	-	-	-	-	-	-	**530**	±75
	Treated discharge	5.2	-	-	3.4	-	-	4.8	-	-	**4.5**	±1.0
#3	Inlet	958	1,140	939	-	-	-	-	-	-	**1,012**	±111
	Control discharge	957	985	1,000	-	-	-	-	-	-	**981**	±22
	Treated discharge	1.9	-	-	0	-	-	11	-	-	**4.4**	±6.1
#4	Inlet	277	242	279	-	-	-	-	-	-	**266**	±21
	Control discharge	334	385	409	-	-	-	-	-	-	**376**	±39
	Treated discharge	3.4	-	-	2.2	-	-	4.6	-	-	**3.4**	±1.2
#5	Inlet	744	769	912	-	-	-	-	-	-	**808**	±91
	Control discharge	622	609	589	-	-	-	-	-	-	**607**	±17
	Treated discharge	0	-	-	0	-	-	0	-	-	**0**	-
#6	Inlet	661	612	638	-	-	-	-	-	-	**637**	±24
	Control discharge	443	526	625	-	-	-	-	-	-	**532**	±94
	Treated discharge	3.9	-	-	0	-	-	2.2	-	-	**2.0**	±1.9

DPM Disintegrations per minute
FR Field replicate
AVG Average
STD Standard deviation

Bacteria

Table B.8 Enumeration of *E. coli*

Test cycle	Water type	*E. coli* (CFU/100 mL)										
		FR1	FR2	FR3	FR4	FR5	FR6	FR7	FR8	FR9	AVG	STD
# 1	Inlet	32	10	10	-	-	-	-	-	-	**17**	±13
	Control discharge	21	10	<10	-	-	-	-	-	-	**14**	±6.4
	Treated discharge	<10	<10	<10	<10	<10	<10	<10	<10	<10	**<10**	-
#2	Inlet	<10	<10	<10	-	-	-	-	-	-	**<10**	-
	Control discharge	43	65	21	-	-	-	-	-	-	**43**	±22
	Treated discharge	<10	<10	<10	<10	<10	<10	<10	<10	<10	**<10**	-
#3	Inlet	32	10	120	-	-	-	-	-	-	**54**	±58
	Control discharge	87	<10	43	-	-	-	-	-	-	**47**	±39
	Treated discharge	<10	<10	<10	<10	<10	<10	<10	<10	<10	**<10**	-
#4	Inlet	10	10	10	-	-	-	-	-	-	**10**	-
	Control discharge	<10	<10	<10	-	-	-	-	-	-	**<10**	-
	Treated discharge	<10	<10	<10	<10	<10	<10	<10	<10	<10	**<10**	-
#5	Inlet	10	<10	21	-	-	-	-	-	-	**14**	±6.4
	Control discharge	<10	<10	<10	-	-	-	-	-	-	**<10**	-
	Treated discharge	<10	<10	10	<10	<10	<10	<10	<10	<10	**<10**	-
#6	Inlet	<10	10	21	-	-	-	-	-	-	**14**	±6.4
	Control discharge	<10	<10	10	-	-	-	-	-	-	**<10**	-
	Treated discharge	<10	<10	<10	<10	<10	<10	<10	<10	10	**<10**	-

CFU Colony-forming units
FR Field replicate
AVG Average
STD Standard deviation

Table B.9 Enumeration of enterococci

Test cycle	Water type	Enterococci (CFU/100 mL)										
		FR1	FR2	FR3	FR4	FR5	FR6	FR7	FR8	FR9	AVG	STD
# 1	Inlet	10	<10	<10	-	-	-	-	-	-	**<10**	-
	Control discharge	10	10	<10	-	-	-	-	-	-	**10**	-
	Treated discharge	<10	<10	<10	<10	<10	<10	<10	<10	<10	**<10**	-
#2	Inlet	<10	<10	<10	-	-	-	-	-	-	**<10**	-
	Control discharge	<10	<10	<10	-	-	-	-	-	-	**<10**	-
	Treated discharge	<10	<10	<10	<10	<10	<10	<10	<10	<10	**<10**	-
#3	Inlet	<10	<10	<10	-	-	-	-	-	-	**<10**	-
	Control discharge	<10	<10	<10	-	-	-	-	-	-	**<10**	-
	Treated discharge	<10	<10	<10	<10	<10	<10	10	<10	<10	**<10**	-
#4	Inlet	<10	<10	<10	-	-	-	-	-	-	**<10**	-
	Control discharge	<10	<10	<10	-	-	-	-	-	-	**<10**	-
	Treated discharge	<10	<10	<10	<10	<10	<10	<10	<10	<10	**<10**	-
#5	Inlet	<10	<10	<10	-	-	-	-	-	-	**<10**	-
	Control discharge	10	<10	<10	-	-	-	-	-	-	**<10**	-
	Treated discharge	10	<10	<10	<10	<10	<10	<10	<10	<10	**<10**	-
#6	Inlet	<10	<10	<10	-	-	-	-	-	-	**<10**	-
	Control discharge	<10	10	10	-	-	-	-	-	-	**10**	-
	Treated discharge	<10	<10	<10	<10	<10	<10	<10	<10	<10	**<10**	-

CFU Colony-forming units
FR Field replicate
AVG Average
STD Standard deviation

Table B.10 Enumeration of *Vibrio cholerae*

Test cycle	Water type	*Vibrio cholerae* (CFU/100 mL)										
		FR1	FR2	FR3	FR4	FR5	FR6	FR7	FR8	FR9	AVG	STD
# 1	Inlet	<1	<1	<1	-	-	-	-	-	-	<1	-
	Control discharge	<1	<1	<1	-	-	-	-	-	-	<1	-
	Treated discharge	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	-
#2	Inlet	<1	<1	<1	-	-	-	-	-	-	<1	-
	Control discharge	<1	<1	<1	-	-	-	-	-	-	<1	-
	Treated discharge	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	-
#3	Inlet	<1	<1	<1	-	-	-	-	-	-	<1	-
	Control discharge	<1	<1	<1	-	-	-	-	-	-	<1	-
	Treated discharge	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	-
#4	Inlet	<1	<1	<1	-	-	-	-	-	-	<1	-
	Control discharge	<1	<1	<1	-	-	-	-	-	-	<1	-
	Treated discharge	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	-
#5	Inlet	<1	<1	<1	-	-	-	-	-	-	<1	-
	Control discharge	<1	<1	<1	-	-	-	-	-	-	<1	-
	Treated discharge	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	-
#6	Inlet	<1	<1	<1	-	-	-	-	-	-	<1	-
	Control discharge	<1	<1	<1	-	-	-	-	-	-	<1	-
	Treated discharge	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	-

CFU Colony-forming units
FR Field replicate
AVG Average
STD Standard deviation

APPENDIX C

QMP and QAPP with Amendments Nos. 1-3

Quality Management Plan

Performance Evaluation of Ballast Water Management Systems

DHI Denmark

Version 2.3

WATER • ENVIRONMENT • HEALTH

September 2011

Quality Management Plan

Performance Evaluation of Ballast Water Management Systems

DHI Denmark
Version 2.3

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Project	Project No.
Quality Management Plan Performance Evaluation of Ballast Water Management Systems DHI Denmark Version 2.3	
Author Gitte I. Petersen	Date 2011.09.07 Approved by Torben Madsen

Revision	Description	By	Checked	Approved	Date
2.3	QMP	GIP	TMA	TMA	7/9-11

Key words	Classification
	☐ Open ☐ Internal ☒ Proprietary

Distribution	No. of copies
Client Certification body DHI: /EAT	

CONTENTS

APPENDICES

1 TERMS AND ABBREVIATIONS

Terms/Abbreviations	Definitions and comments
Active substance	A substance which has a general or specific action on aquatic organisms or bacteria (pathogens)
Ballast Water Management System (BWMS)	A system which removes, renders harmless or avoids uptake or discharge of aquatic organisms and bacteria (pathogens) with ballast water and sediments by mechanical, physical, chemical or biological means acting individually or in combination
Certification Body	Body to certify facilities to conduct performance evaluation of BWMS according to the IMO Convention
Client	The party requesting a performance evaluation of a technology.
Convention	The IMO convention on ballast water
International Maritime Organization (IMO)	United Nations specialized agency with responsibility for the safety and security of shipping and the prevention of marine pollution by ships Comment: IMO has adopted the International Convention for the Control and Management of Ship's Ballast Water and Sediments
Quality Assurance Project Plan (QAPP)	Project-specific technical document describing the BWMS to be tested, the test facility and other conditions affecting the actual design and implementation of the required experiments
Quality Management Plan (QMP)	Generic document describing the quality control management structure and policies of the testing body (including subcontractors and outside laboratories)
Services	The performance evaluation of maritime technologies by laboratory, land-based or shipboard tests or a combination hereof
Standard Operation Procedure (SOP)	Generic document providing rules, guidelines or characteristics for tests or analyses Comment: In-house methods may be used in the absence of a recognized standard, if they are commonly accepted for testing of BWMS or scientifically documented

2 INTRODUCTION

The International Maritime Organization (IMO) has adopted the International Convention for the Control and Management of Ship's Ballast Water and Sediments /1/ to reduce the risk of spreading of harmful aquatic organisms and pathogens released with ballast water.

The Convention requires that all ships comply with specified water quality requirements (D2) before ballast water is released into the environment.

The performance evaluation of ballast water management systems (BWMS) aims at documenting compliance with the requirements stated in international guidelines, e.g.:

- Guideline for approval of ballast water management systems - G8 /2/

- Procedure for approval of ballast water management systems that make use of active substances - G9 /3/.

DHI provides services in relation to performance evaluation of maritime technologies and particularly BWMS within the DHI Ballast Water Centre which includes test facilities and laboratories in Denmark and Singapore.

The DHI Ballast Water Centre is organized with a Ballast Water Facility Board including two members from the management in DHI Denmark and two members from the management in DHI Singapore. The object of the Board is to coordinate the development and marketing of services related to the performance evaluation of BWMS within the DHI Group.

The services addressed with the present Quality Management Plan (QMP) include:

- Laboratory tests conducted at the DHI environmental laboratory in Hørsholm, Denmark, aiming at proof-of-concept or technology optimisation
- Pilot-tests conducted at the DHI Maritime Technology Evaluation Facility (hereafter referred to as the "test facility") in Hundested, Denmark, aiming at technology optimisation
- Land-based tests conducted at the test facility according to international guidelines
- Shipboard tests conducted by DHI Denmark according to international guidelines at vessels, on which the technology is installed

The above activities are collectively referred to as the "services" whereas individual activities are referred to as "projects".

The aim of the services is to provide independent, third party documentation for the performance of maritime technologies. High quality of the services is ensured through extensive quality management and use of skilled staff.

3 ORGANISATION

3.1 Head of department (Torben Madsen)

The head of department, business strategy, for DHI's Department of Environment and Toxicology, has the overall responsibility for the services and the test facility. This includes the following tasks:

- Co-ordination of joint business development between DHI Denmark and DHI Singapore via participation in the Ballast Water Facility Board
- Negotiation of agreements (i.e. service contracts) with clients
- Responsibility for overall co-ordination, planning and costs as required to ensure that the appropriate human resources, facilities and equipment are available for the services
- Appointment of the business area manager, the project manager and task leaders for cross-cutting functions (e.g. production of test water and test facility technical operations)
- Maintenance of the QMP with updated versions as appropriate

- Approval of the Quality Assurance Project Plan (QAPP) and Standard Operation Procedures (SOPs)
- Quality control and approval of test reports (provided that the head of department has not contributed to the technical solution of the project)
- Documentation in relation to
    - Staff training and experience
    - Facilities and their maintenance
    - Records of complaints

3.2 Business area manager (Gitte I. Petersen)

The business area manager is responsible for the scientific and technical quality of the services in co-ordination with the head of department. This includes the following managerial tasks:

- Business development and marketing
- Maintenance of generic standards that can serve as formats for drafting the QAPPs and approval of the methods applied in land-based and shipboard tests
- Dialogue with task leaders for cross-cutting functions, e.g. production of test water and test facility technical operations
- Contributions to data interpretation and reporting of land-based and shipboard tests in collaboration with the project manager
- Participation in discussions with the Certification Body on important matters, particularly draft and final reports, together with the project manager
- Co-ordination of the services with the aim to ensure feasibility of parallel projects conducted at the test facility, including decisions related to the functioning of the test facility (e.g. piping and pumps)
- Maintenance of the test facility, connection piping between the test facility and the client's technology, and dialogue with academic and technical staff in order to fulfil DHIs responsibility for operating the test facility during testing
- Quality control of test reports (provided that the business area manager has not contributed to the technical solution of the project)

3.3 Project manager

The project manager is responsible for the management and efficient performance of the project in accordance with the contract between the client and DHI, the QMP and the QAPP.

The project manager's tasks include:

- Organisation and management of the project
- Periodic meetings and other communication with the client to ensure that all necessary information is available in due time
- Preparation of the draft and final QAPP with detailed description of the project, including time schedule and quality assurance of deliverables

- Facilitation of the process for comments and responses to the draft QAPP in dialogue with the client and the Certification Body
- Preparation of amendments and deviations to the QAPP, if any
- Communication of the project time schedule to the Certification Body to enable external audit
- Communication of the QAPP and project time schedule to the internal auditor identified in the QAPP to enable internal audit
- Participation in discussions with the Certification Body on important matters, particularly draft and final reports, together with the business area manager
- Co-ordination and dialogue with the business area manager in relation to safe conditions of work, logistics and technical operations at the test facility
- Co-ordination and dialogue with the laboratory manager in relation to the practical organisation of work involving laboratory technicians; the project manager shall in due time inform the laboratory manager on the types of tests and the required capacity to enable laboratory capacity planning
- Agreements with subcontractors as appropriate for meeting the project deliverables (e.g. chemical analytical laboratory)
- Approval of initiation of the test cycles and interruption of test cycles, e.g. in case of irregularity
- Preparation of reports

3.4 Head of projects

The academic staff (with exception of the business area manager, project manager, task leaders for cross-cutting functions and test co-ordinators) and the secretaries are appointed by the head of projects via dialogue with the business area manager or the project manager as appropriate.

3.5 Laboratory manager

The laboratory manager appoints laboratory technicians for a specific project and allocates tasks to them as part of the laboratory capacity planning. Furthermore, the laboratory manager appoints one or more test co-ordinators among the laboratory technicians or the academic staff for on-site co-ordination of land-based test cycles.

3.5.1.1 Academic staff, laboratory technicians and secretaries

The tasks of the academic staff, the laboratory technicians and the secretaries include:

- Contributions to the QMP, QAPP and SOPs
- Test co-ordinator function, i.e. co-ordination and keeping timely records of the activities at the test facility during land-based tests
- Sampling at the test facility
- Monitoring of test water quality
- Maintenance of materials and equipment
- Analysis and data processing
- Contributions to test reports

- Archiving of documents and raw data

4 PERFORMANCE OF PROJECT

4.1 Agreement

An agreement between the client and DHI is negotiated and signed according to the DHI manual for project management.

4.2 Quality Assurance Project Plan (QAPP)

The QAPP is a project specific document describing the technology to be tested, the test facility, and other conditions affecting the actual design and implementation of the study. The QAPP is only required for performance evaluation of BWMS in land-based or shipboard tests conducted according to international guidelines.

The QAPP is

- Prepared by the project manager
- Signed by the project manager, the head of department and the internal auditor from the DHI Quality Assurance Unit
- Forwarded to the Certification Body for review and comments
- Forwarded to the client for review, acceptance and signature.

The QAPP typically includes the following titles:

1. Objective
2. Client (including client's monitor, if any)
3. Administration
4. DHI Ballast Water Centre
5. Subcontractors
6. Project management
7. System description
8. Safety handling of active substances
9. Test design (including, for **land-based test**, test cycles, test water, sampling and analyses, and, for **shipboard test**, trial period and locations, sampling and analyses)
10. Validity criteria
11. Pass criteria
12. Time schedule
13. Quality assurance
14. Report
15. Archiving
16. Amendments and deviations, if any
17. References

The QAPP refers to a number of SOPs (see Appendix A).

Amendments and deviations to the QAPP are approved and signed by the project manager. Amendments describe planned changes whereas deviations describe unplanned changes to the QAPP.

The QAPP is subject to internal audit in accordance with the procedures for internal audit of the DHI Quality Management System.

4.3 Services

The project will be conducted as described in the QAPP and subsequent amendments and deviations or, alternatively, as described in the agreement between the client and DHI for projects, for which no QAPP is prepared.

4.3.1 Laboratory tests

Laboratory tests can be initiated when the BWMS technology is ready for testing and DHI's deliverables are defined. Initiation of testing is decided by the project manager in agreement with the client.

4.3.2 Pilot tests

Pilot tests can be initiated when the BWMS technology is installed and ready for operation. Initiation of testing is decided in consensus by and between the business area manager and the project manager in agreement with the client.

4.3.3 Land-based tests

Land-based tests can be initiated when the BWMS technology is installed and ready for operation. Initiation of testing is decided in consensus by and between the business area manager and the project manager in agreement with the client.

The project manager decides when a test cycle in the land-based test is completed and valid, when appropriate by reference to the G8 guidelines /2/, G9 guidelines /3/ or other standards (e.g. US requirements). If required, the project manager can decide to interrupt a test cycle due to technical malfunctioning of the test facility or the BWMS, insufficient state of biological or physical parameters or for other reasons related to the quality of the test water.

4.3.4 Shipboard tests

Shipboard testing can be initiated when the BWMS technology is installed on the vessel and ready for operation. Initiation of testing is decided by the project manager in agreement with the client.

The project manager decides when a test cycle in the shipboard test is completed and valid by reference to the criteria in G8 /2/ or, if appropriate, to criteria in other standards (e.g. US requirements). If required, the project manager can decide to interrupt a test cycle due to technical malfunctioning of the BWMS, insufficient state of biological or physical parameters or for other reasons related to the water quality.

4.4 Reports

Reports are prepared with the details, format and language described in the agreement between the client and DHI.

4.4.1 Performance evaluation of BWMS under the IMO convention

For land-based or shipboard tests of BWMS conducted as part of the IMO approval process, the report is typically structured by use of the appropriate headings in the QAPP and shall include a summary of any amendments and deviations to the QAPP.

The report shall include all relevant technical and analytical data and will contain at least the following items:

- Name and address of the client (and monitor, if any)
- Name and address of the testing facility and the dates, on which the test was initiated and completed
- Objectives and procedures stated in the approved QAPP including any changes made to the QAPP
- Results obtained, presented in summarizing tables and as raw data
- Any unforeseen circumstances which may have affected the quality or integrity of the land-based/shipboard testing
- Storage locations of all raw data, the signed QAPP and report
- Descriptions of operations, calculations and transformations performed on the presented data
- Quality assurance statement

The report shall be signed by the project manager, the internal auditor from the DHI Quality Assurance Unit and the head of department.

The final report will be prepared in English and forwarded to the client.

5 QUALITY MANAGEMENT PROCESSES

5.1 DHI Quality Assurance

The services are conducted in accordance with the principles of ISO 9001 by using the DHI Quality Manual and the procedures in this QMP. The Quality Management System of DHI is found compliant with ISO 9001 as part of the ISO 17025 accreditation of the DHI environmental laboratory.

The DHI quality manager is responsible for assigning a trained internal auditor from DHI's Quality Assurance Unit to each project in accordance with the procedures for internal audit of the DHI Quality Management System.

The internal auditor is identified in the QAPP. The internal auditor shall receive the QAPP from the project manager in order to plan and execute internal audit of the project.

5.2 Document and record control

The DHI Quality Manual includes a procedure describing the process of drafting, revising and approving documentation. Standard operation procedures are controlled as described in SOP 30/944.

SOPs 30/921 and 30/937 describe how records of the test are stored, transferred, maintained and controlled in order to ensure data integrity for a period defined in the QAPP, but not shorter than 5 years from completion of the verification.

5.3 Internal audits

Procedure 3 in the DHI Quality System Manual on audit and evaluation and SOP 30/943 describe the process of periodic internal auditing of projects and activities including audit responsibilities and planning, auditor training and competences and audit reporting.

Procedure 4 in DHI Quality System Manual on non-conformities and corrective actions describes how deviations identified during operation and auditing are corrected (corrective actions) and how future occurrence of the same deviations is prevented by improving the quality manual including the process descriptions and working methods (preventive actions).

5.4 Complaint management

Procedure 5 in the DHI Project Management Manual on Complaints describes how complaints are recorded, resolved and reported. If not resolved, complaints are referred to the Certification Body for resolving.

5.5 Subcontractor management

Procedure 4 in the DHI Project Management Manual on subcontractors describes how it is ensured that subcontractors follow quality requirements.

In addition, analytical laboratories providing analyses of any kind should:

- Maintain an ISO 17025 accreditation with the quality management system required herein.
- Apply accredited analytical methods when available.
- Apply other methods according to either international standard methods or in-house methods that are in all cases validated as required for accredited methods.

SOP 30/700 furthermore describes how it is ensured that purchased items such as chemicals and glassware are controlled, accepted and calibrated.

5.6 Staff competence management

Procedure 3 on appraisal interview, post qualifying education and experience in the DHI Employee Conditions Handbook describes how it is ensured that the projects are conducted by staff with adequate competences and knowledge. This is done by maintaining a list of functions in the test process with competence requirements and responsibilities. The list is supported by reference to staff files in the DHI CV database.

5.7 Facility management

SOP 30/945 describes how it is ensured that facilities and equipment are available and fit for the purposes.

5.8 Management review

Procedure 3 of the Quality System Manual on audit and evaluation describes how the DHI management is ensuring that the test centre is working according to this quality manual through mechanisms such as e.g. an annual management review process.

The Quality Manager is responsible for maintenance and development of the quality system and for the internal auditing of all aspects of the system – with daily reference to the Director, Group R&D and Quality Management. The DHI Quality Manual contains rules for reviews of the quality system.

6 REFERENCES

/1/ IMO (2005): International Convention for the Control and Management of Ships Ballast Waterand Sediments. London. International Maritime Organization.

/2/ MEPC. Guidelines for approval of ballast water management systems (G8). resolution MEPC.174(58). Adopted 10th October 2008.

/3/ MEPC. Procedure for approval of ballast water management systems that make use of active substances (G9). MEPC.126(53) Adopted 22nd July 2005.

APPENDIX A

BMWS testing-specific Standard Operating Procedures (SOPs)

SUBJECT/SUBSUBJECT	NO.
ANALYTICAL METHOD ZOOPLANKTON ANALYSIS	30/1700:04
ANALYTICAL METHOD MICROSCOPIC ENUMERATION AND IDENTIFICATION OF MICROALGAE (LUGOL AND CMFDA/FDA)	30/1701:02
ANALYTICAL METHOD DETERMINING PRIMARY PRODUCTION OF MICROALGAE	30/1702:03
ANALYTICAL METHOD DETERMINING DIVERSITY OF MICROALGAL COMMUNITIES BY HPLC ANALYSIS OF PIGMENTS	30/1703:03
ANALYTICAL METHOD DETERMINATION OF VIABLE ALGAE BY MPN	30/1704:02
MICROBIOLOGICAL TESTS DETERMINATION OF TOTAL NUMBER OF BACTERIA BY EPIFLUORESCENCE MICROSCOPY	30/1705:03
MICROBIOLOGICAL TESTS DETERMINATION OF HETEROTROPHIC PLATE COUNT	30/1706:03
MICROBIOLOGICAL TESTS DETERMINATION OF *VIBRIO CHOLERAE* IN WATER	30/1707:02
MICROBIOLOGICAL TESTS DETERMINATION OF TOTAL COLIFORM, *E.COLI* AND ENTEROCOCCI Colilert*-18 AND Enterolert-E	30/1708:02
MEASUREMENT METHOD OZONE MEASUREMENT IN WATER	30/1730:02
MEASUREMENT METHOD OZONE MEASUREMENT IN AIR	30/1731:02
MEASUREMENT METHOD TRO MEASUREMENT IN WATER	30/1732:02
HARVESTING, CULTURING AND ADDITION OF ORGANISMS	30/1734:03
COLLECTION OF SEAWATER	30/1735:02
COLLECTION OF FRESH WATER	30/1736:02
CHEMICAL CRITERIA FOR TEST WATER ADDITION OF DOC, POC AND MM	30/1737:02
SAMPLING BIOLOGICAL AND WATER QUALITY PARAMETERS	30/1738:02
SAMPLING WET TEST	30/1739:02
SAMPLING DBP ANALYSIS	30/1740:02
STATISTICS STATISTICAL SURVEILLANCE OF BIOLOGICAL DATA OBTAINED AT TESTS OF BWMSs	30/1760:01
LABELLING SAMPLES COLLECTED AT TEST SITE	30/1761:01
OPERATION OF THE DHI MTEF	30/1762:02
CLEANING RETENTION TANKS, PIPINGS AND OTHER EQUIPMENT AT TEST SITE	30/1763:02
MEASUREMENT METHOD ON-LINE MONITORING OF PRESSURE, TEMPERATURE AND FLOW RATES AT TEST SITE	30/1764:01
MEASUREMENT METHOD FLUORESCENCE	30/1765:02

SUBJECT/SUBSUBJECT	NO.
MEASUREMENT METHOD TURBIDITY	30/1766:03
HEALTH AND SAFETY ENSURING WORKER HEALTH AND SAFETY AT TEST SITE	30/1767:02
MEASUREMENT METHOD DETERMINATION OF TSS	30/1768:02
MEASUREMENT METHOD DETERMINATION OF DOC AND POC	30/1769:02

A P P E N D I X   B

Overview of lists

Overview of lists

The lists mentioned below are kept together with the rest of quality documentation.

Certification Body

DHI holds a statement describing the Certification Body that has certified the DHI Maritime Technology Evaluation Facility.

List of sub-contractors

DHI keeps a list of sub-contractors used during the test. The list contains information on name of company, address, contact person, e-mail, telephone number and deliveries.

List of staff approved for functions at the test facility

DHI keeps a list of persons working at the test facility. The list contains information on the person's activities, responsibility and documentation for training. The person's competence is documented in an available CV.

List of Standard Operation Procedures

DHI keeps a list of SOPs, including those used in relation to projects conducted at the test facility.

APPENDIX C

Template for amendments to QAPP

AMENDMENT

QAPP DOCUMENT TITLE AND DATE:

AMENDMENT NUMBER:

DATE OF AMENDMENT:

AMENDMENT CONTENTS:

REASON FOR AMENDMENT:

IMPACT OF AMMENDMENT:

PREVENTATIVE ACTION:

If relevant, action to prevent that the same cause of amendment will reoccur in the future.

ORIGINATED BY:

SIGNED BY:

____

Project manager

____

DATE

Copy to be sent to the client, the Certification Body and the DHI Quality Assurance Unit.

A P P E N D I X   D

Template for amendments to QAPP

DEVIATION

QAPP DOCUMENT TITLE AND DATE:

DEVIATION NUMBER:

DATE OF DEVIATION:

DESCRIPTION OF DEVIATION:

REASON FOR DEVIATION:

IMPACT OF DEVIATION:

CORRECTIVE ACTION:

If required, actions to be taken to prevent consequences of deviation

ORIGINATED BY:

SIGNED BY:

____
Project manager

____
DATE

Copy to be sent to the client, the Certification Body and the DHI Quality Assurance Unit.

Quality Assurance Project Plan

Shipboard Test of Auramarine's Crystal Ballast 250 Ballast Water Management System

October 2011

Quality Assurance Project Plan

Shipboard Test of Auramarine's Crystal Ballast 250 Ballast Water Management System

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Client	Client's representative
Auramarine Ltd.	Jukka Suvanto

Project	Project No
Shipboard Test of Auramarine's Crystal Ballast 250 Ballast Water Management System	11811494
Authors	**Date:**
Michael Andersen	2011-10-21
	Approved by
	Torben Madsen

Revision	Description	By	Checked	Approved	Date
		MJA	LSC	TMA	
1	First draft	MJA	LSC	TMA	2011.10.21

Key words	Classification
	☐ Open ☐ Internal ☒ Proprietary

Distribution	No of copies
Auramarine Ltd. DNV DHI	Electronic

CONTENTS

APPENDICES

1 OBJECTIVE

For an application for final approval, the IMO Convention requires an approval of Ballast Water Management Systems (BWMS) according to the principles laid down in Resolution MEPC.174(58) (G8) /1/ to assure that BWMS approved by administrations are capable of meeting the standard regulation D-2 (MEPC G8) in land-based and shipboard evaluations and do not cause unacceptable harm to the vessel, crew, environment or public health.

The objective is to conduct a shipboard test of the Auramarine Crystal Ballast 250 BWMS (hereafter CB250) according to Resolution MECP.174(58), Guidelines for approval of ballast water management systems (G8) (hereafter designated as the 'G8 guidelines').

2 CLIENT

Auramarine Ltd.
P.O. Box 849
FIN-20101 Turku
Finland

Contact person: Jukka Suvanto

3 CERTIFICATION BODY

Det Norske Veritas AS
Veritasveien 1
N-1363 Høvik, Oslo
Norway

4 DHI BALLAST WATER CENTRE

DHI
Agern Allé 5
DK-2970 Hørsholm
Denmark

Contact Person: Michael Jakob Andersen

5 SUBCONTRACTORS

The shipboard test will be conducted by DHI, and, with the possible exception of verification of *Vibrio cholerae* (according to SOP 30/1707), subcontractors will not be involved.

6 PROJECT MANAGEMENT

The project manager for the study is Michael Jakob Andersen.

7 SYSTEM DESCRIPTION

The description of the CB250, provided in Appendix A, is identical to the description received from the client.

8 SAFETY HANDLING OF ACTIVE SUBSTANCES

No active substances are applied by the BWMS, CB250.

9 TEST DESIGN

9.1 Trial period and locations

The shipboard test will be conducted during at least two separate campaigns on board the vessel M/S Ahtela, IMO 8911736, DNV ID 16627. The campaigns will be conducted within a trial period with a time span of not less than six months.

The first campaign (Campaign 1) will include two test cycles, one by use of a Boll&Kirch 6.18.2 filter and one by use of a FilterSafe BSF-100H filter which are connected to the BWMS (as described in Appendix A). Campaign 1 is scheduled to be conducted in Hundested, Denmark, between 28.10.2011 and 08.11.2011. The time of arrival of M/S Ahtela in Hundested shall be communicated to DNV and DHI by email from Auramarine. The testing locations are due to change if the densities of phytoplankton in the sea in Hundested are below the validity criteria for inlet water described in section 10.

The second campaign (Campaign 2) will include two test cycles by use of one of the above-mentioned filters, which will be selected after Campaign 1. Campaign 2 will be conducted in April-May 2012 or later. Details on dates and locations for ballasting and deballasting activities will be provided as amendments to the Quality Assurance Project Plan when this information is available.

9.2 Test cycles

The BWMS will be operated by Auramarine during all the test cycles. Each test cycle consists of sampling and analyses of:

Inlet water (the physico-chemical and biological parameters in the inlet water will be considered as sufficiently stable during the ballasting; unless the local conditions indicate that the parameters in the inlet water vary with time, only one set of samples and analyses will be used to represent the control tank and the ballast tank);

Discharge control water (stored without treatment from the time of ballasting to discharge);

Discharge treated water (treated and stored from the time of ballasting to discharge).

9.3 Sampling

9.3.1 Sample overview

	Samples	**Sample volumes per replicate**
Inlet water	3 replicates	Organisms ≥50 µm: >1 m^3 *
		Organisms 10-50 µm: >1 L **
		Bacteria: >0.5 L **
		DOC + POC: approx. 0.5 L **
		TSS: approx. 2 L **
Discharge control water	3 replicates	Organisms ≥50 µm: >1 m^3 *
		Organisms 10-50 µm: >1 L **
		Bacteria: >0.5 L **
		DOC + POC: approx. 0.5 L **
		TSS: approx. 2 L **
Discharge treated water	3 replicates	Organisms ≥50 µm: >3 m^3 *
	3 x 3 replicates	Organisms 10-50 µm: >1 L **
	3 x 3 replicates	Bacteria: >0.5 L **
	3 replicates	DOC + POC: approx. 0.5 L **
	3 replicates	TSS: approx. 2 L **

*, collected by continuous flow during the entire period of intake or discharge; this continuous sampling of 3 replicates, each with a volume of at >3 m^3, provides the same statistical basis for counting as the sampling 3 x 3 replicates of >1 m^3 which is recommended in the G8 guidelines; **, grab samples collected over the period of intake or discharge (e.g. start, middle and end).

9.3.2 Samples for enumeration of organisms ≥50 µm

Three replicates will be collected by parallel continuous sampling during the entire periods of intake and discharge. The samples will be gently filtered through a net with a mesh size of 35 µm and a reservoir (cod-end) at the bottom of the net for collecting the zooplankton. Each replicate will be concentrated in 1-L glass bottles. The total volume of the filtered sample will be determined by a flow meter.

9.3.3 Samples for enumeration of organisms ≥10 µm and <50 µm

Grab samples (3 replicates of the inlet water, 3 replicates of the control discharge water, and 3 x 3 replicates of the treated discharge water) with a volume of at least 1 L will be collected in appropriate containers.

9.3.4 Samples for enumeration of bacteria

Grab samples (3 replicates of the inlet water, 3 replicates of the control discharge water, and 3 x 3 replicates of the treated discharge water) with a volume of at least 0.5 L will be collected in appropriate sterile containers.

9.4 Analyses

9.4.1 Analysis overview

	Temperature	Salinity	≥ 50 µm	10-50 µm, Lugol's	10-50 µm, MPN	10-50 µm, Primary production	Bacteria	DOC + POC	TSS
	Replicates								
Inlet water									
Replicate 1 (start)	1	1	Three replicates	1	1	1	1	1	1
Replicate 2 (mid)	2	2		2	2	2	2	2	2
Replicate 3 (end)	3	3		3	3	3	3	3	3
Control discharge water									
Replicate 1 (start)	1	1	Three replicates		1	1	1	1	1
Replicate 2 (mid)	2	2			2	2	2	2	2
Replicate 3 (end)	3	3			3	3	3	3	3
Treated discharge water									
Replicate 1-3 (start)	1	1	Three replicates		1-3	1	1-3	1	1
Replicate 4-6 (mid)	4	4			4-6	4	4-6	4	4
Replicate 7-9 (end)	7	7			7-9	7	7-9	7	7

The samples for all analyses will be kept cool from the time of collection, and the samples will be analysed within shortest possible time period. The following sections use the wording "work on location" to describe activities carried out on-board the vessel. In the case that the shipboard test is conducted on M/S Ahtela alongside the pier in Hundested, it may be convenient to collect samples on-board and perform the analyses in DHI's laboratory in Hørsholm (approx. 1 hour drive from Hundested). "Work on location" will in this situation have no meaning as all analyses will be conducted in the laboratory.

9.4.2 Temperature and salinity

Temperature and salinity will be measured by use of portable multi parameter instrument equipped with electrodes. Measurements will be conducted at regular intervals throughout the inlet and discharge operations.

9.4.3 Organism size class ≥50 µm

The concentrations of viable organisms ≥50 µm in the samples will be determined by using a stereo microscope and a counting chamber according to SOP 30/1700. Viable organisms will be determined on the basis of mobility and morphology and by using the vital stain Neutral Red. The viable organisms will be characterized according to broad taxonomic groups such as rotifers, crustaceans, molluscs, worms, etc. The analyses will be completed on location.

9.4.4 Organism size class ≥10 µm and <50 µm

Work on location. *Inlet water samples* will be treated with Lugol's solution to enable determination of the concentrations of organisms in the size class ≥10 and <50 µm. The container with the inlet water sample (approx. 10 L) will be shaken gently (upside down 5 times); two subsamples (approx. 100 mL) per replicate will be transferred immediately to brown 100-mL glass bottles and added Lugol's solution to achieve 2% final concentration according to SOP 30/1701.

The concentrations of viable algae in the *inlet and discharge water samples* will be analyzed by use of the most probable number (MPN) assay. The container with the total sample (approx. 10 L) will be shaken gently (upside down 5 times). For the MPN assay, dilution series of the inlet water, control discharge water and treated discharge water will be prepared by adding aliquots of sample to test tubes with liquid medium to a total volume of 6 mL, including controls containing only 6 mL of medium, as described in SOP 30/1704. All test tubes will be kept at ambient temperature without direct exposure to the sun, where after the incubation will be continued upon arrival to the laboratory.

For measuring the primary production of algae in *inlet and discharge water samples*, two representative subsamples of each replicate will be transferred to 60 mL bottles and incubated for approx. 2 hours under light from a light-panel. After incubation, the samples will be filtered onto GF/D filters and the filters will be transferred to glass vials as described in SOP 30/1702.

Work in laboratory. The concentrations of viable organisms in the size class ≥10 and <50 µm in the inlet water will be determined by *inverted microscopy* of samples preserved with Lugol's solution according to SOP 30/1701. The analyses comprise detailed examination of the algal chloroplasts to confirm that the phytoplankton were alive and classification of the algae in major taxonomic groups.

Most probable number (MPN) assay. Upon arrival to the laboratory, the fluorescence of the test tubes will be determined immediately. The MPN test tubes will be incubated for

14 days at room temperature as described in SOP 30/1704. The concentrations of viable algae in the inlet water, control discharge water and treated discharge water will be determined by measuring of the fluorescence in the MPN test tubes according to SOP 30/1704.

Primary production will be determined by measuring the amounts of ^{14}C fixed by photosynthesis by liquid scintillation counting according to SOP 30/1702.

9.4.5 Bacteria

Work on location. The concentrations of *E. coli* and enterococci will be determined according to SOP 30/1708 with appropriate modifications for shipboard test. The analyses of *E. coli* and enterococci will be completed on location when the sampling time allows sufficient time for incubation and enumeration. If there is not time to complete the analyses on location, one sample (volume approx. 100 mL) per replicate will be transferred to sterile containers which will be kept in the dark at 1-4 ºC until the arrival at the DHI laboratory.

Samples for detection of *Vibrio cholerae* will be filtered through a 0.45 µm-filter, where after the filter will be placed in a sterile container which will be kept in the dark at 1-4 ºC until the arrival at the DHI laboratory.

Work in laboratory. Analyses of *E. coli* and enterococci that were not completed on location will be conducted in the laboratory according to SOP 30/1708.

The possible occurrence of *Vibrio cholerae* will be analysed according to SOP 30/1707 with appropriate modifications for shipboard test.

9.4.6 DOC, POC and TSS

Work on location. For determination of dissolved organic carbon (DOC) and particulate organic carbon (POC), an appropriate sample volume is treated as described in SOP 30/1769.

For determination of total suspended solids (TSS) an appropriate sample volume is filtered through a glass fibre filter which has already been weighed in the laboratory.

Work in laboratory. Determination of DOC and POC according to SOP 30/1769. Determination of TSS according to SOP 30/1768.

10 VALIDITY CRITERIA

Valid test cycles are test cycles in which:

- the concentrations of viable organisms in the inlet water are at least 10 times higher than the maximum permitted values in regulation D-2.1 on discharge (excepted from the requirements to bacteria);

- the concentrations of viable organisms in the discharge control water exceed the maximum permitted values in regulation D-2.1 on discharge (excepted from the requirements to bacteria).

Organism size class	IMO D-2.1 requirements on discharge
Organisms ≥ 50 µm	<10 viable organisms/m^3
Organism size: ≥10 µm - < 50 µm	<10 viable organisms/mL

11 PASS CRITRIA

The G8 guidelines prescribe that the performance evaluation in the shipboard test may be considered successful, if the BWMS has passed the criteria below in three consecutive test cycles, including ballasting and deballasting operations, conducted on board a vessel during a trial period of not less than six months.

The pass criteria for the shipboard test cycles are:

1. The test cycle shall be valid according to the validity criteria
2. The average density of organisms larger than or equal to 50 µm in minimum diameter in the replicate samples shall be less than 10 viable organisms per m^3 at discharge
3. The average density of organisms smaller than 50 µm and larger than or equal to 10 µm in minimum diameter in the replicate samples shall be less than 10 viable organisms per mL at discharge
4. The average density of *Vibrio cholerae* (serotypes O1 and O139) shall be less than 1 CFU per 100 mL at discharge
5. The average density of *E. coli* in the replicate samples shall be less than 250 CFU per 100 mL at discharge
6. The average density of intestinal enterococci in the replicate samples shall be less than 100 CFU per 100 mL at discharge

12 TIME SCHEDULE

October-November 2011	First campaign of test cycles conducted on board
April-May 2012	Second campaign of test cycles conducted on board
May-June 2012	Draft and final reporting. Final report submitted one month after completion of Campaign 2

13 QUALITY ASSURANCE

The DHI Quality Assurance Unit will review this Quality Assurance Project Plan and conduct inspections of the laboratory analyses and the raw data.

The final report will be audited.

Inspection and audit will be carried out by Quality Assurance personnel independent of the staff involved in the shipboard test.

14 REPORTS

The following reports will be prepared:

An interim report compiling the data for the first campaign of test cycles

A draft final report compiling all relevant data from the test cycles, data interpretation and conclusion

A final report

15 ARCHIVING

All data generated and all other records and information relevant to the quality and integrity of the land-based testing will be retained according to the DHI Quality Manual Plan /2/. The data will be filed in the archives of DHI and retained for a period of five years after issue of the final report.

16 AMENDMENTS AND DEVIATIONS

Amendments are planned changes of the Quality Assurance Project Plan. Deviations are unplanned changes. Amendments and deviations will be signed by the project manager and documented in the file and the final report.

17 REFERENCES

/1/ Resolution MEPC.174(58). Adopted on 10 October 2008. Guidelines for approval of ballast water management systems (G8).

/2/ Quality Management Plan (QMP) for DHI Maritime Technology Evaluation Facility (MTEF). 2010.

APPROVAL OF QUALITY ASSURANCE PROJECT PLAN

DHI Ballast Water Centre

Project management	Michael Andersen Michael Jakob Andersen	Date: 21/10-11
DHI management	Torben Madsen	Date: 21/10-11
Quality Assurance Unit	Louise Schlüter	Date: 21/10-11

This QAPP is accepted and my signature authorises the study to proceed as described in this document.

Client	Jukka Suvanto Auramarine Ltd.	Date:

A P P E N D I X   A

Description of the ballast water management system as given by the client

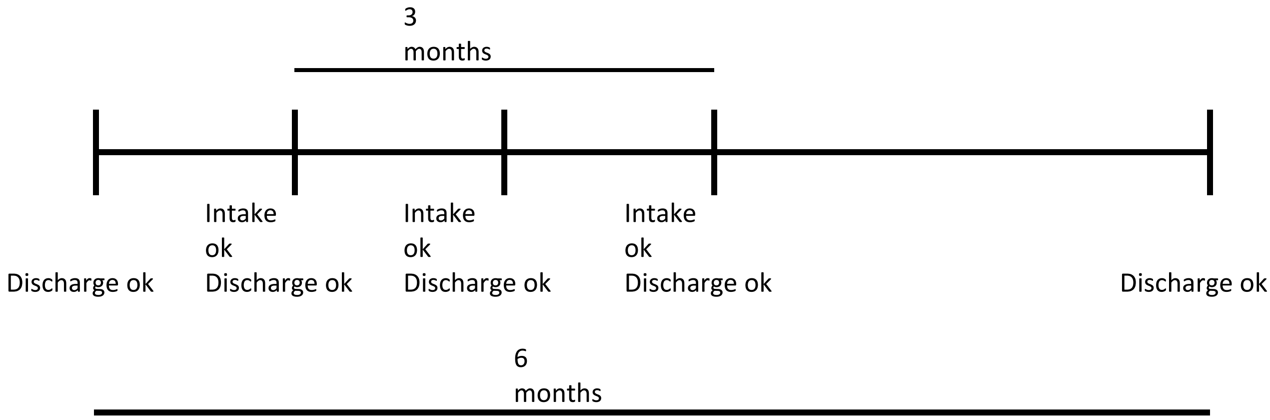
SHIPBOARD TEST PLAN ver.1.2
CrystalBallast® CB75/250
BALLAST WATER TREATMENT SYSTEM
ONBOARD M/S AHTELA, IMO 8911736, DNV ID 16627
AURAMARINE

Document Revision 1.0

Dated: 14/10/2011

Jukka Suvanto

Auramarine has a ballast water treatment system called CrystalBallast. Auramarine will perform shipboard tests according to IMO G8 in ship called Ahtela. Testing will be done for two sizes of CrystalBallast 75m³/h (CB75) and 250m³/h (CB250). Testing period will be for CB250 6 months of discharge fulfilling the G8 recommendations, and it includes a 3 month period where the intake and discharge are fulfilling the G8 recommendations. As in the timeline below.



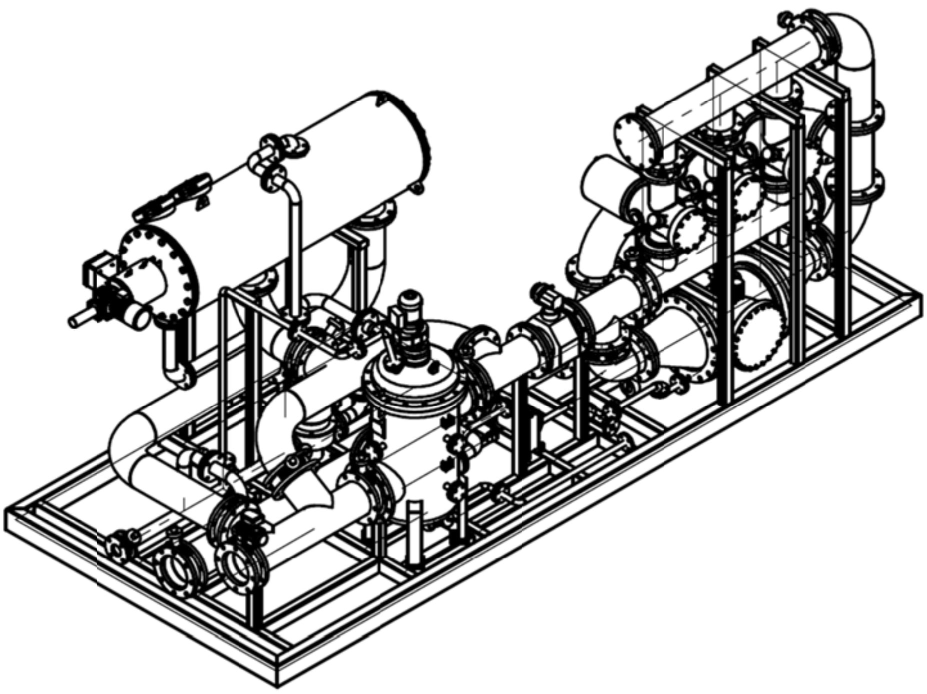
The CB75 unit will be tested for 3 months where the intake and discharge will fulfill the G8 recommendations.

The ship Ahtela is not in any regular route. The G8 is recommending that the ship should be in normal operation. Because Ahtela is not in any regular route she will simulate a normal operation by ballasting/deballasting 3-4 times per week every other week. Also the ship will sail between two ports between the ballasting/deballasting. The ports can be any ports in the world. This is considered as normal operation. If Ahtela will have some ‘sold’ routes during the 6 month period, this will be considered as normal operation.

CrystalBallast® CB 75/250 Combo for testing purposes onboard M/S AHTELA

Auramarine Ltd manufactures for testing purposes a combined 75 m3/h + 250 m3/h CrystalBallast® ballast water treatment (BWT-) unit, which will be installed in two 20' containers; BWT filter units and UV-reactors in one container and the power and control units in another container, ref. Auramarine's drawing nr 308421. The unit includes one Boll&Kirch 6.18.2 filter and one FilterSafe BSF-100H (BSFc-H/V-16) filter. The two filters are installed to the same unit to ensure Auramarine can achieve the Type Approval. Auramarine has had some problems with the Boll&Kirch filter. If the problems continue Auramarine will perform the tests with the FilterSafe filter.

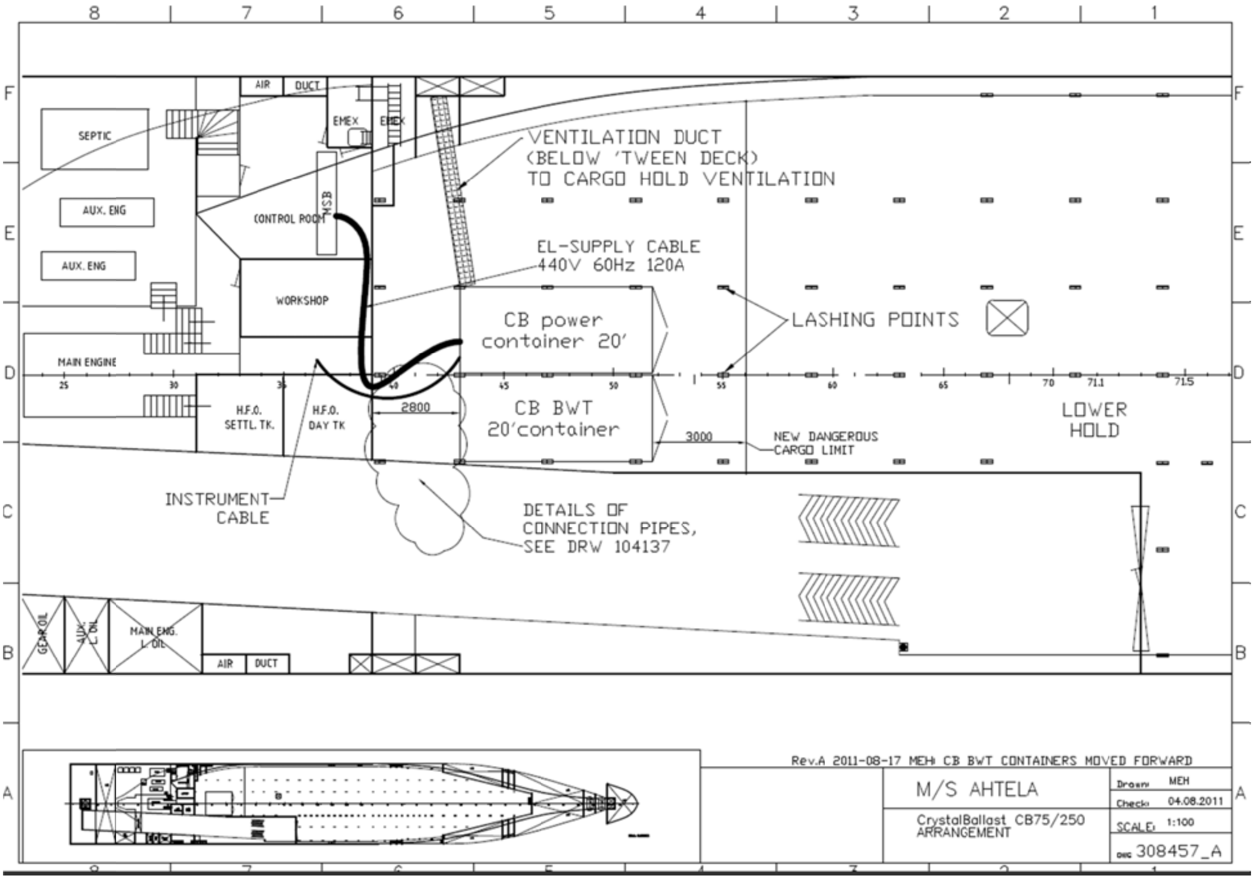
The unit also has two different kinds of UV-reactors. For CB250 there is one piece of CR-A250 reactor and for CB75 there is three pieces of CR-A75 reactors. There is three pieces of reactors to meet the pump capacity of the Ahtela's ballast pumps. Basically Auramarine will perform two tests with 225m3/h (3*75m3/h) and 250m3/h. The units PI is in the appendix 1.



Picture 1: GA of the unit

Arrangement of installation

The containers will be installed in ship's lower hold, aft part in the vicinity and corner of ramp and aft bulkhead towards engine room, see drawing nr 308457 A "CrystalBallast CB75/250 Arrangement on M/S AHTELA".

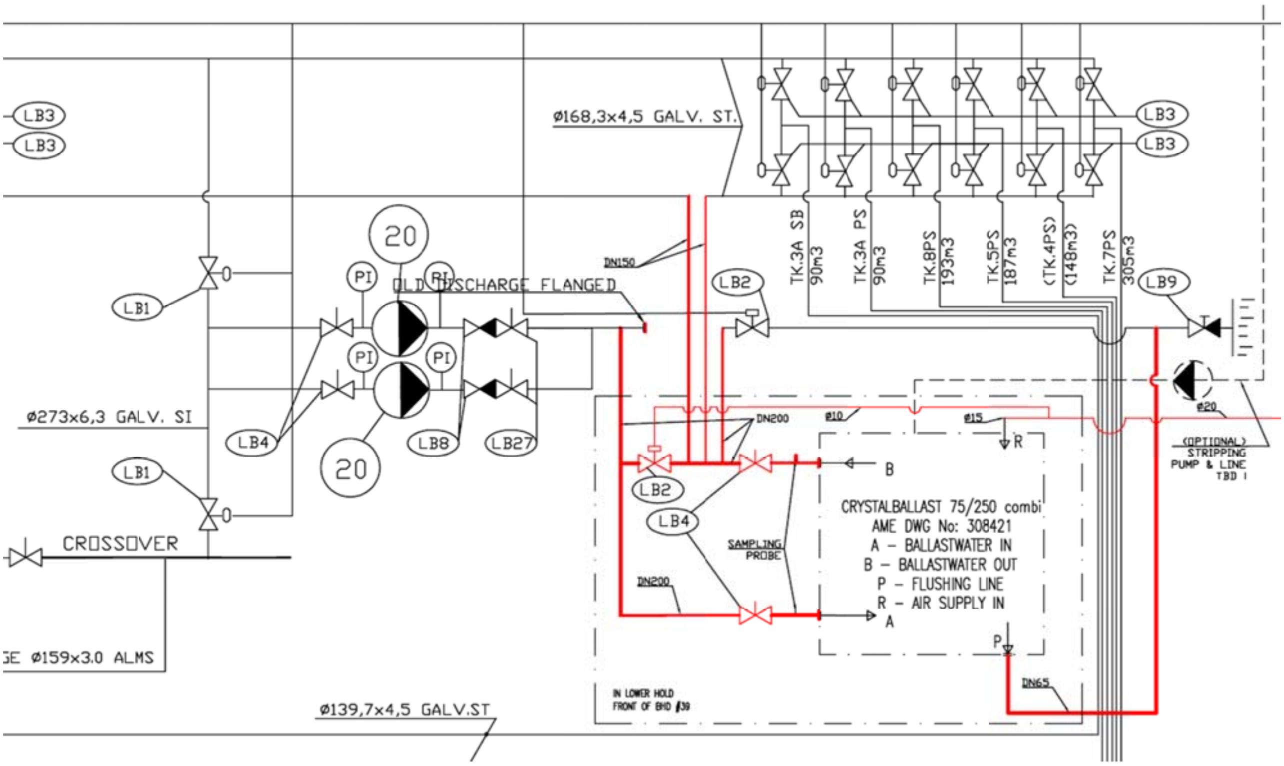


Picture 2: position of containers

Picture 3: view of the aft lower cargo hold

Connection to existing ballast system

Connections to ships bilge- and ballast system will be done according to rev.D3 of diagram 1-051-0016 D3_(BILGE AND BALLAST SYSTEM)



Picture 4: PI of the ship ballasting system

Sample water overflow from BWT-unit will be led by using movable hoses to lower hold bilge well, S-side aft corner, and further to (cargo hold) bilge discharge.

In the diagram there is shown an optional change of stripping system; this change will not be applied in this phase, yet, and the existing ejector system remains onboard. Thus requirements stated in DNV Pt.6 Ch.18 Sec.4 B206 will not be fulfilled, but it does not effect on test equipment's performance or testing procedures. From the ship's point of view, the changes on the stripping system become actual when the IMO BW-Code comes in to force.

The following changes and additions on ship's piping will be made:

1. Remote controlled system by-pass valve DN200; “LB2”
2. Closing of old ballast discharge line with flange, moving of remote operated discharge valve and connection to new discharge line from BWT unit
3. new connection pipes to BWT unit (ZnSt), DN 200 and to ballast system DN150; through bulkhead #39
4. 2 x manually operated closing valves DN200 between BWT unit and ship’s BW system (to be open normally)
5. Flushing line pipe DN65 from BWT-unit to existing side discharge valve. During the filter back flush valve LB2 is closed. This prevents the untreated water to enter the tanks.
6. Control air supply pipe, dia 18mm, from ship’s instrument air dryer

7. Sampling pipes, see drawing 308456, in cargo space between the containers and machinery space, in order to make the sampling & testing easy.
8. During ballasting the valve LB2 on the ballast discharge line will be closed. By closing this valve the system will avoid the possibility of filter back flush to be returned to main line.

Sampling points

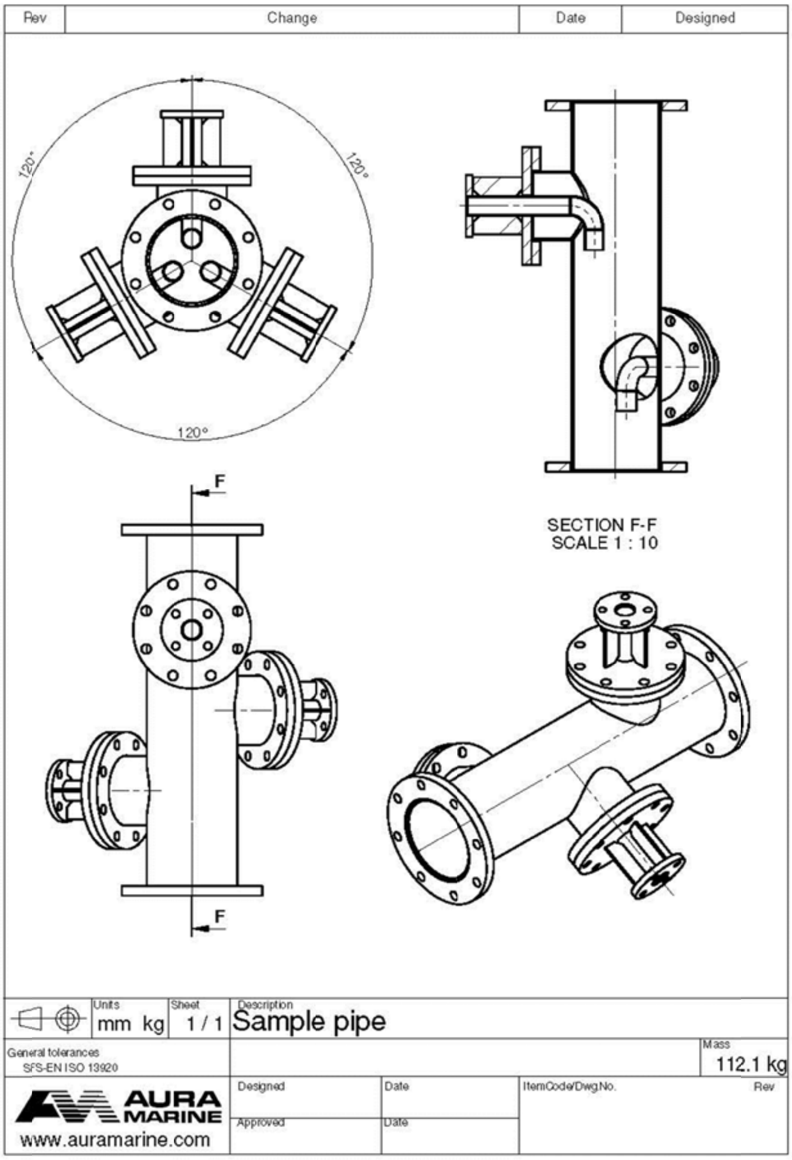
On the system inlet and outlet there will be sample points which are recommended in the G2 and G8. There will be 3 pieces of sample points on the system inlet and outlet, totally 6 pieces of sampling points. To have multiple sampling points Auramarine wants to confirm that there will be enough samples and that the sample volumes are large enough.

The sample points are in the same cargo space as the containers which include the unit. This is because there is not space for sampling equipment in the engine room. The sample points are as in picture 5. The sampling points are located in the pipeline before and after the unit as in picture 6.

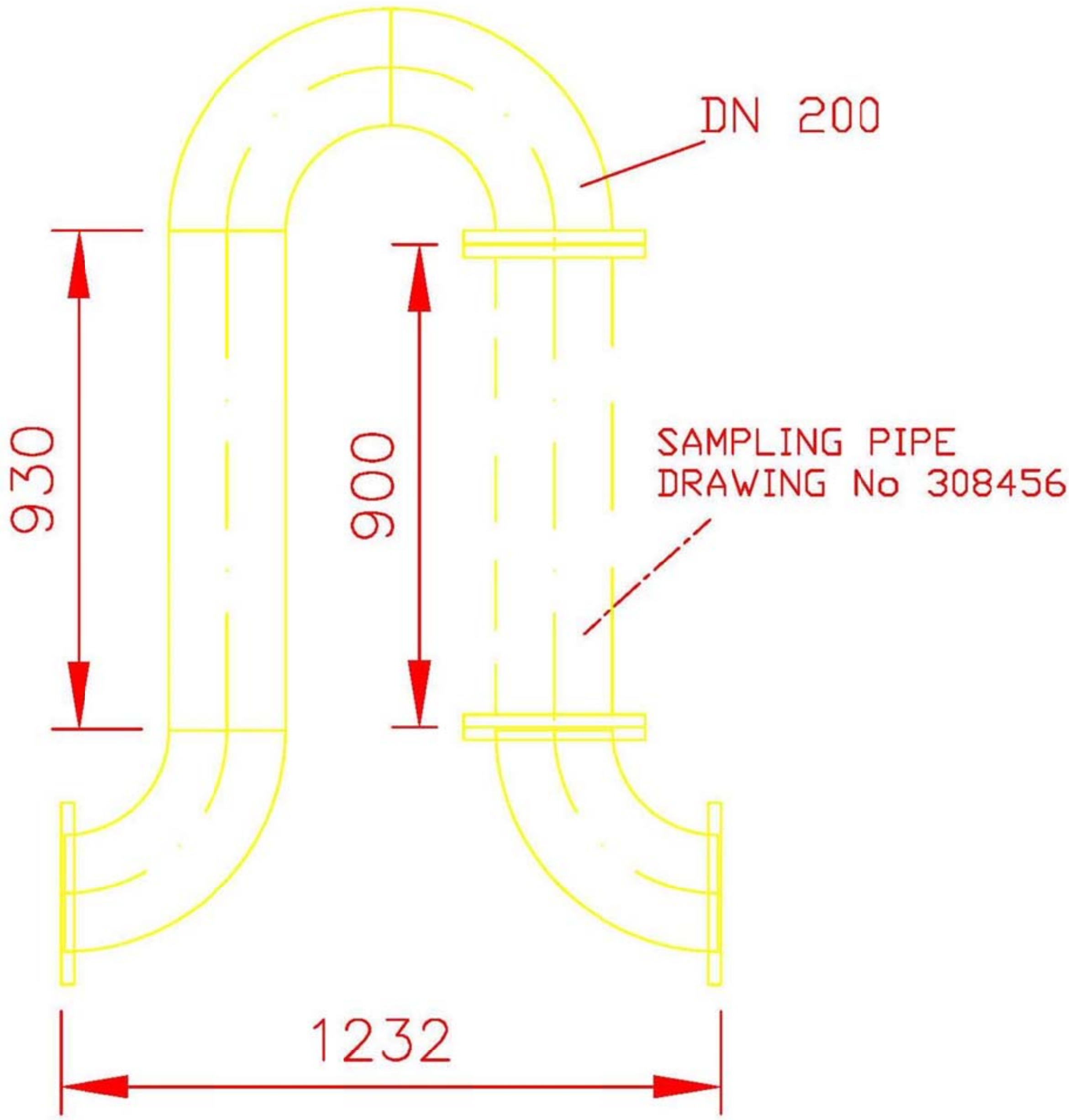
DNV comment: For the sampling points, please note that placing the 2 sample outlets as proposed may influence the samples of the 2nd outlet, depending on the direction of the flow. The QAPP for shipboard tests should indicate how the samples will be taken and from which outlet. In any case, it will be an interesting study to see if turbulence caused by the first sampling outlet will affect the sampling results from the 2nd sampling outlet.

Testing plan

Auramarine is aiming to perform the tests starting on October 2011 and ending on April 2012. For the shipboard tests Ahtela will be sailed to the seas where there will be enough of phytoplankton to meet the G8 recommendations. This will probably be in North Sea outside of Holland etc. If there will not be enough phytoplankton, Ahtela will sail more South. Auramarine will perform the CB75 and CB250 tests at the same time. Only difference is that the tests for CB75 will last at least 3 months and for CB250 tests will last at least 6 months.



Picture 5: Sample pipe



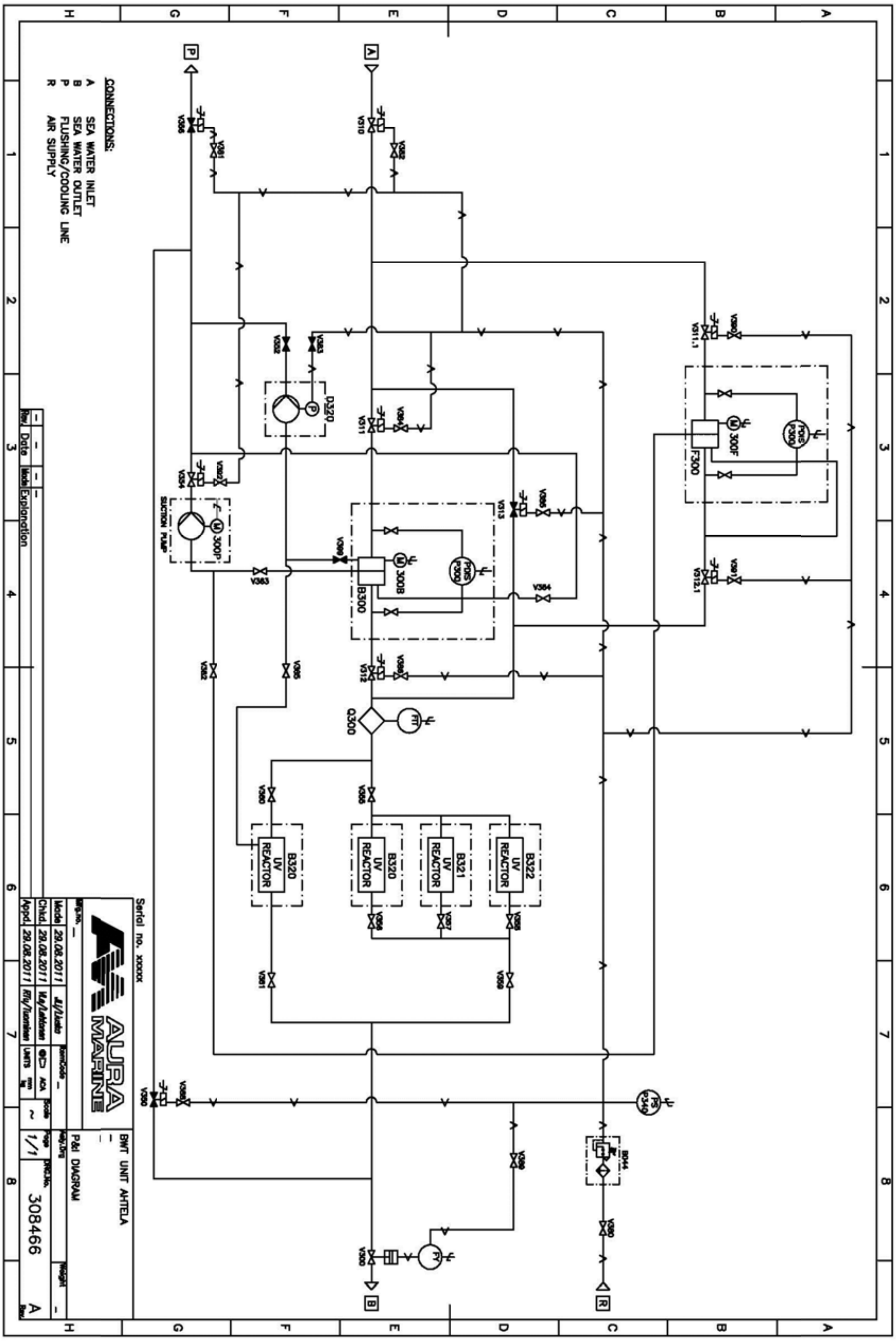
Picture 6: Pipeline on the sampling points

By performing the shipboard tests according to this document Auramarine will have conducted shipboard tests required by the G8 Guidelines and BWM.2/Circ.33 as part of Type Approval.

The Type Approval is given to CrystalBallast models CB75 and CB250. The Type Approval also allows Auramarine to build units with bigger flows by installing a filter which can handle bigger flow and installing the UV-reactors in parallel.

Other shipboard tests will also be continued in MS Pasila. In Pasila the test continues on CB500 and CB1000. If the 6 month shipboard test will be conducted earlier in Pasila than Ahtela, this will give Type Approval for CB250, CB500 and/or CB1000.

If then again the 6 month test period will be conducted first in Ahtela on CB250. Then other models CB75, CB500 ja CB1000 will be tested for 3 months.


CONNECTIONS:
A SEA WATER INLET
B SEA WATER OUTLET
P FLUSHING/COOLING LINE
R AIR SUPPLY
AURA MARINE
BWT UNIT AHTELA
P&I DIAGRAM
308466
UV REACTOR
B320
B321
B322
B300
F300
Q300
SUCTION PUMP
Serial no. xxxxx

Appendix 1.

AMENDMENT

Quality Assurance Project Plan

Shipboard Test of Auramarine's Crystal Ballast 250 Ballast Water Management System. October 2011.

Amendment number 1

2011.10.28

Amendment Comments

The CrystalBallast® CB75/250 Combo

The QAPP in general applies for the shipboard testing of the Auramarine CrystalBallast® CB75/250 combo ballast water treatment unit and not only the CB250 as described in the QAPP. The descriptions in the QAPP are also valid for the CB75. The test design and analyses will be exactly the same for the CB75 and the CB250.

The first campaign (Campaign 1) with the CB75 will include two test cycles, one by use of a Boll&Kirch 6.18.2 filter and one by use of a FilterSafe BSF-100H. Campaign 1 is scheduled to be conducted in Hundested, Denmark, between 05.11.2011 and 09.11.2011. The M/S Ahtela is currently situated in Hundested. The testing location is due to change if the densities of phytoplankton in the sea in Hundested are below the validity criteria for inlet water described in section 10 of the QAPP.

Amendment to section 9.4.1 Analysis overview.

Enumeration and characterization of organisms in the ≥10 µm and <50 µm size range by inverted microscopy will also be conducted for control discharge water samples fixed with Lugol solution. This will be conducted in addition to MPN assays for discharge control water. The methodology will be the same as described for inlet water control samples in section 9.4.4 of the QAPP.

Reason for Amendment

Due to the potential favourable conditions for shipboard testing in Hundested, Denmark Auramarine Ltd. has requested to initiate testing with the CB75 during the planned testing campaign for the CB250 in October-November 2011.

The addition to section 9.4.1 analysis overview of the QAPP was made according to a request from DNV.

Impact of Amendment

The trial period of at least three months for the CB75 described in Appendix A of the QAPP is planned to be initiated on the 5th of November 2011.

Preventive Action

Not relevant.

Michael Andersen

Michael Andersen

____

Project manager

2011.10.28

Date

Copy to be sent to the client, the Certification Body and the DHI Quality Assurance Unit.

AMENDMENT

Quality Assurance Project Plan

Shipboard Test of Auramarine's Crystal Ballast 250 Ballast Water Management System. March 2012.

Amendment number 2

09.03.2012

Amendment Comments

The second campaign (Campaign 2) with the CrystalBallast75 (CB75) will include two test cycles with a Boll&Kirch 6.18.2 filter. Campaign 2 is scheduled to be conducted in Hundested, Denmark, between 12.03.2012 and 16.03.2012. The M/S Ahtela is scheduled for arrival in Hundested 11.03.2012. The testing location is due to change if the densities of phytoplankton in the sea in Hundested are below the validity criteria for inlet water described in section 10 of the QAPP.

Reason for Amendment

Planned amendment with details on locations and dates for Campaign 2 as described in section 9.1 of the QAPP.

Impact of Amendment

The trial period of the CB75 will be completed by the 16.03.2012.

Preventive Action

Not relevant.

Michael Andersen

Michael Andersen

____
Project manager

09.03.2012
Date

Copy to be sent to the client, the Certification Body and the DHI Quality Assurance Unit.

AMENDMENT NO. 3

Quality Assurance Project Plan

Shipboard Test of Auramarine's Crystal Ballast 250 Ballast Water Management System

April 2012

Date

18.04.2012

Amendment

The second campaign (Campaign 2) with the CrystalBallast250 (CB250) will include two test cycles; one with a FilterSafe BSF-100H filter and one with a Boll&Kirch 6.18.2 filter. Campaign 2 is scheduled to be conducted in Hundested, Denmark, between 28.04.2012 and 01.05.2012. The testing location is due to change if the densities of phytoplankton in the sea in Hundested are below the validity criteria for inlet water described in section 10 of the QAPP.

Reason for Amendment

Planned amendment with details on locations and dates for Campaign 2 with the CB250 as described in section 9.1 of the QAPP.

Impact of Amendment

The trial period of the CB250 will be completed by the 01.05.2012.

Preventive Action

Not relevant.

Michael Andersen

____

Michael Andersen, Project manager

Copy to be sent to the client, the Certification Body and the DHI Quality Assurance Unit.

APPENDIX D

Certificate of compliance, ISO 9001 certificate, accreditation and GLP authorisation

COPY

Lloyd's Register Certificate of Compliance

Office: **Lloyd's Register EMEA**
Copenhagen Design Support Centre, Statutory Section
Strandvejen 104A, 2nd floor
DK-2900 Hellerup
Denmark

Date: **09 May 2012**

This certificate is issued to **DHI Ballast Water Centre, Denmark**

DHI Ballast Water Centre, Denmark

The Document(s) listed in paragraph 1 of the appendix have been examined for compliance with:

- Resolution MEPC.174(58), Annex part 2

and are found to comply from quality assurance and quality control aspects subject to the following:

1.1. It is required to maintain full and accurate log files in order to demonstrate correct quality measures

1.2. The Quality Assurance Project Plan is a project specific document and should as such be subject to review and commenting prior to each project start-up.

1.3. This design appraisal document is to be kept together with quality management plan.

1.4. Subject certificate is valid until 15 June 2015.

1. The documents listed below have been examined

Drawing No.	Rev.	Title	Status	Date
Date: 07 Sep 2011	**2.3**	**Quality Management Plan**	**B**	**09 May 2012**

2. The documents listed below have been considered together with the submitted documents in the appraisal

Drawing No.	Rev.	Title
11810704	**02**	**Quality Assurance Project Plan**

Appraisal Status Key

B Examined and found to comply with §2.2, Part 2 of the annex of IMO Resolution MEPC 174 (58)

Martin Schabert
Statutory Department
Copenhagen Design Support Centre
Surveyor to Lloyd's Register EMEA

LLOYD'S REGISTER EMEA
Lloyd's Register
M. Schabert
COPENHAGEN DESIGN SUPPORT CENTRE

A member of the Lloyd's Register Group

DET NORSKE VERITAS

MANAGEMENT SYSTEM CERTIFICATE

Certificate No. 109333-2012-AQ-DEN-DANAK

This is to certify that

DHI Group

has been found to conform to the management system standard:

DS/EN ISO 9001:2008

This certificate is valid for the following product or service ranges:

Consulting, software, research & development and laboratory testing, analysis & products within the area of water, environment & health

Locations included in the certification will appear in the appendix.

Place and date:

Hellerup, 2012-01-10

DET NORSKE VERITAS,
BUSINESS ASSURANCE, DANMARK A/S

This certificate is valid until:

2015-01-10

The audit has been performed under the supervision of:

DANAK
SYSTEM Reg.nr. 5001

Jan Carsten Schmidt
Lead Auditor

Jens Peter Høiseth
Managing Director

Lack of fulfilment of conditions as set out in the Certification Agreement may render this certificate invalid.

DET NORSKE VERITAS

APPENDIX TO CERTIFICATE

This appendix refers to certificate no. 109333-2012-AQ-DEN-DANAK

DHI Group

Locations included in the certification are as follows:

Site Address	Scope:
Agern Allé 5 2970 Hørsholm, Denmark	Consulting, MIKE© by DHI Software Development, Sales & Support, Solutions Software Development, Research, Development & Innovation and Laboratory Analysis, Testing & Products
INCUBA Science Park, Gustav Wieds Vej 10 8000 Århus, Denmark	Consulting, Solutions Software Development and Research, Development & Innovation

Place and date:

Hellerup, 2012-01-10

DET NORSKE VERITAS,
BUSINESS ASSURANCE, DANMARK A/S

This certificate is valid until:

2015-01-10

The audit has been performed under the supervision of:

DANAK
SYSTEM Reg.nr. 5001

Jan Carsten Schmidt
Lead Auditor

Jens Peter Høiseth
Managing Director

Accreditation to testing

COPY DANAK

Company: **DHI**

Agern Allé 5
DK-2970 Hørsholm

Registration number: 26

Valid: **04-07-2011 to 31-07-2015**

Scope:

Testing

Product

- **Biological items for testing**
- **Chemicals, chemical products, cosmetics, fertilizers, paints**
- **Environmental samples: Air, water, soil, waste**
- **Construction products**

Test Type

- **Biological, biochemical testing**
- **Chemical testing, Analytical chemical testing**
- **Radiochemistry, radiation**
- **Sampling, laboratories accredited for sampling**

Testing is performed according to the current list of test methods approved by DANAK.

The company complies with the criteria in EN ISO/IEC 17025:2005 – General requirements for the competence of testing and calibration laboratories and demonstrates technical competence for the defined scope and the operation of a quality management system (refer joint ISO-ILAC-IAF Communiqué dated January 2009, www.danak.dk).

Issued July 4th 2011

Jesper Høy / **Kirsten Jebjerg Andersen**

In case of any disputes, the Document in Danish language shall have priority

COPY

DANAK

GOOD LABORATORY PRACTICE

STATEMENT OF COMPLIANCE

Laboratory inspection and study audits for compliance with the OECD Principles for Good Laboratory Practice were carried out at

Laboratory: DHI

on date: 25th March 2010 plus 7th and 9th April 2010

The laboratory inspection and study audits have been carried out in accordance with the regulation settled in Order No. 906 of 14th September 2009 from the Danish Ministry of Environment. The laboratory has been monitored for GLP Compliance within the following scope:

Type of products:

- *Industrial chemicals*
- *Pesticides*
- *Biocides*

Type of tests:

- *Environmental toxicity studies on aquatic and terrestrial organisms.*
- *Studies of behaviour in water, soil and air, bioaccumulation*

The laboratory was found to be operating in compliance with the OECD Principles of Good Laboratory Practice.

Date: 2nd December 2010

Jesper Høy
Managing director, DANAK

Kirsten Jebjerg Andersen
GLP inspector, DANAK